Attorney Docket No. 5218-39C PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re: Anagnostou et al.
Serial No.: 09/525,808
Filed: March 15, 2000
For: *METHOD OF TREATING ENDOTHELIAL INJURY*

Confirmation No. 9764
Examiner: Christopher H. Yaen
Group Art Unit: 1642

October 27, 2005

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION UNDER 37 C.F.R § 1.132
OF GEORGE SIGOUNAS, Ph.D.

Sir/Madam:

I, George Sigounas, Ph.D., do hereby declare and say as follows:

1. I received my Ph.D. from Boston University in Cellular Biology. I am currently Professor of Medicine at East Carolina University School of Medicine in Greenville, North Carolina. A *curriculum vitae* is attached herewith at Tab 1. I am a co-inventor on the above-identified patent application.

2. I have reviewed the Office Actions, dated August 10, 2004 and December 28, 2004, respectively, in the above-captioned patent application and am familiar with the contents thereof. I have also reviewed Silvestris et al. *Ann Hematol.* 70(6): 313-318 (1995), de Vos et al. Cancer Treat Rev. 30(6): 495-513 (2004), JP 02 096535 to Chugai Pharmaceutical Co. Ltd. and Bukowski et al. *Blood* 84(2)(Supp. 1):129a (1994) cited in the Office Actions.

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3. One of my research interests relates to the study of erythropoietin as a protective agent of the endothelium. The endothelium plays an important role in angiogenesis reproduction, embryonic vasculogenesis, wound healing, blood clotting, and in many serious diseases, such as atherosclerosis and cancer. Far from being a passive vessel lining, endothelial cells upon exposure to environmental agents (e.g. chemotherapeutic drugs, radiation, chemicals, heat, etc.), biological factors (e.g. cytokines, chemokines, growth factors, LPS, infections, diseases, etc.) or mechanical means (e.g. physical trauma, balloon injury, surgery, etc.) undergo profound structural and functional alterations and in several occasions are irreparably damaged. This damage can lead to disturbance of vital functions such as hemostasis, inflammatory reactions and immunity.

Erythropoietin (EPO) is a 30.4 KD glycoprotein involved in the proliferation, differentiation and survival of erythroid cells. It is produced primarily in the kidneys. EPO is indicated for the treatment of anemia.

EPO has been used to treat anemia in cancer patients. Current clinical guidelines for treatment of cancer patients with anemia include the following:

- Use Epoetin in patients with chemotherapy-associated anemia with a hemoglobin (Hgb) concentration below 10g/dL;
- Use Epoetin subcutaneously thrice weekly for a minimum of 4 weeks;
- In the absence of response, do not continue Epoetin beyond 6-8 weeks;
- Titrate Epoetin once Hgb reaches 12g/dL or restart it when the level falls to near 10g/dL; and
- For anemic patients with hematologic malignancies, initiate conventional therapy and observe hematologic response before considering use of Epoetin.

Prior to September 11, 1996, it was not common practice to administer EPO to anemic cancer patients prior to receiving treatment with a chemotherapeutic agent. Instead, anemic cancer patients who received EPO, were administered EPO only after undergoing chemotherapy and after being diagnosed with anemia. Attached at Tab 2 is a copy of the prescribing information for Procrit® (Epoetin alpha, recombinant human

erythropoietin) from the 1996 Physician's Desk Reference (PDR) (50th ed. 1996). The prescribing information for Procrit® indicates that Procrit® is administered to anemic cancer patients undergoing chemotherapy. Accordingly, the prescribing information indicates that Procrit® is administered to patients receiving chemotherapy, and not administered to patients prior to diagnosis of anemia or prior to treatment with a chemotherapeutic agent.[1]

Additionally, attached at Tab 4 are studies published in 1993 (Case et al.) and 1994 (Cascinu et al.) that further show that treatment protocols for anemic cancer patients did not include administration of EPO prior to administration of chemotherapy. Instead, patients previously diagnosed with cancer and receiving chemotherapy were diagnosed as having anemia and were subsequently administered EPO. One aspect of the present invention is directed to the timing of administration of EPO prior to the administration of chemotherapy or other agents and/or events known to cause endothelial injury.

4. We hypothesized that EPO protects and/or repairs endothelial cells and the endothelium from toxicity induced by chemotherapeutic agents. Accordingly, experiments designed to test the *in vivo* effects of administering EPO prior to or in conjunction with a chemotherapeutic drug were carried out under my direction. In particular, the study presented below is directed toward investigating the effects of EPO on structural and functional changes in endothelial cells and the endothelium caused by bleomycin.

To perform the *in vivo* studies, we employed the following protocol outlined below.

[1] Attached at Tab 3 is a copy of the 2005 prescribing information for Procrit®, as of this 2005 publication, the prescribing information for Procrit® still does not indicate the administration of EPO prior to administration of a chemotherapeutic agent for anemic cancer patients, or any patient population.

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Animals

Female C57Bl mice, 6-8 weeks old and weighing 15 to 20 grams were used and treated as it is shown in the following protocol.

Treatment Protocol

Groups: **EPO**, injected with EPO alone

CONTROL, injected with phosphate-buffered saline (PBS)

BLEO, treated with Bleomycin alone

EPO/BLEO treated with EPO and Bleomycin (EPO given prior to administration of BLEO, and EPO and BLEO subsequently injected simultaneously)

Animals: 4-8 per group

Injections: ip, x2 per week

Agents: EPO (5-40 u/mouse)

Bleomycin (0.25-0.75 u/mouse)

Sacrificed: 2 wks, 4 wks, 6 wks

Note: Epo1 (40 u/mouse); Epo2 (10 u/mouse)

Histology

Animals were deeply anesthetized by ketamine/xylazine for sacrifice. The lungs were removed for macroscopic and microscopic analysis. Lung specimens were routinely fixed in 10% neutral buffered formalin and embedded in paraffin. Five-micrometer thick sections were placed on slides and stained with hematoxylin and eosin (H&E), Masson's trichrome stain or factor VIII for qualitative and quantitative morphometric analysis. Cellular alterations of the endothelium were determined by blind analysis of the lung sections using a Nikon microscope.

ICAM-1 Immunostaining and Image Analysis

All samples were routinely processed with 10% formalin and embedded in paraffin. Four-micrometer thick sections from the tissue blocks were placed on PLUS slides. After air drying, tissue sections were deparaffinized and rehydrated before immunostaining. Antigen retrieval was performed by enzyme pretreatment and heating of the tissue sections in a steamer. ICAM-1 specific monoclonal antibody was directed at the extracellular domain of ICAM-1 antigen and detection of bound antibody was achieved using a commercial kit from Santa Cruz Biotechnology Inc.

Expression of intercellular adhesion molecule-1 (ICAM-1) by endothelial cells was quantified using a Nikon microscope equipped with the SPOT-Advanced image-analysis system and assessing 20 to 30 randomly selected fields via grid analysis. A total of 100 endothelial cells were counted and expressed as positive or negative for immunostaining.

Results

Under normal circumstances, the endothelial cells (EC) are spindle-shaped elongated cells. Endothelial cells form the endothelium in the vascular system. The endothelium is a single cell layer separating the lumen of the vessels from the interstitial space. A lung section stained with H&E, as shown in the figure at Tab 5 (panel A), shows a vessel with normal endothelium and endothelial cells. The normal endothelium controls the trafficking of fluids, macromolecules, micromolecules and cells into and out of the lumen. However, when the endothelial cells are activated by various means, including chemotherapeutic agents, they become round and are called prominent endothelial cells. The figure at Tab 5 (panel B) shows another lung section stained with H&E presenting a vessel with interrupted endothelium and prominent (activated) endothelial cells. The prominent endothelial cells form a discontinuous endothelium, homeostasis is disturbed and a series of events takes place, which results in tissue damage.

A. Suppression of endothelial cell activation induced by bleomycin

We investigated the structural changes in the endothelium of the lungs caused by bleomycin in animals treated with the drug alone and in combination with EPO at various time intervals as described in the treatment protocol. Lungs obtained from animals treated with various concentrations of EPO or saline had very low levels of prominent endothelial cells, indicating that this treatment does not stimulate the endothelium. *See* Figure at Tab 6. Sections of lungs collected from animals injected with bleomycin alone displayed approximately 400% more prominent endothelial cells than those found in lungs collected from mice treated with EPO alone or saline ($P<0.05$). A 6-fold decrease in prominent endothelial cells was found in lung sections obtained from mice treated with bleomycin and EPO compared to the animals injected with bleomycin alone ($p<0.008$).

B. Suppression of endothelial cell intercellular adhesion molecule-1 (ICAM-1) expression induced by bleomycin

In our studies, two weeks following treatment, the endothelium of bleomycin-treated animals expressed high levels of intercellular adhesion molecule-1 (ICAM-1). *See* figure at Tab 7; bars represent means $\pm$ SE. In 26 assessed fields, 67 $\pm$ 11.9 % of the endothelial cells were positive for ICAM-1. However, only 25 $\pm$ 6.4 % of lung endothelial cells derived from animals treated with bleomycin and EPO expressed ICAM-1. Thus, EPO induced a 2.7-fold suppression of adhesive molecule expression ($P=0.009$). A linear relationship between the total cumulative dose of bleomycin and the severity of endothelial cell alterations was seen. While low concentrations of bleomycin had no significant effect on the adhesive molecule over the first two weeks, ICAM-1 expression increased at four weeks following treatment.

These results suggest that the ability of EPO to protect the endothelium from drug-induced toxicity is mediated by suppressing ICAM-1 expression and inhibiting endothelial cell activation.

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Our studies indicate that EPO can prevent, protect, and/or repair endothelial damage. This damage may be caused by chemotherapeutic agents and/or events that damage the endothelium structure and/or function. The discovery of such means to protect against endothelial injuries or enhance damage recovery will be beneficial for subjects and may decrease the morbidity and/or mortality rate for conditions associated with endothelial injury.

6. I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

George Sigounas
George Sigounas, Ph.D

10/27/05
Date

CURRICULUM VITAE

GEORGE SIGOUNAS, MS,Ph.D.
April 2005

PERSONAL INFORMATION

Name:	GEORGE SIGOUNAS
Home Address:	313 Queen Anne's Road Greenville, NC 27858 (252) 355-2172
Business Address:	East Carolina University Brody School of Medicine Department of Internal Medicine Hematology/Oncology Division Brody Building, Room 3E-102 Greenville, NC 27858-4354 252-744-2560
Birth date:	June 26, 1954
Birthplace:	Ioannina, Greece
Marital Status:	Married, two children

EDUCATION

University of Patras, Greece - B.Sc. (Biology-Chemistry), 1976.

Northeastern University, Boston, MA, M.S. (Physiology-Biology), 1982.

Boston University, Boston, MA, Ph.D. (Cell Physiology-Biology), 1987.

EMPLOYMENT HISTORY

Professor of Medicine, Hematology/Oncology Division, Department of Medicine, School of Medicine, East Carolina University, Greenville, NC, 1998 - Present.

Director, Stem Cell Processing/Bioengineering Unit, East Carolina University School of Medicine, Department of Medicine, Hematology/Oncology Section, Sept. 1994 - Present.

Adjunct Professor of Biology, Department of Biology, East Carolina University, 1998-Present.

Associate Professor of Medicine, Hematology/Oncology Division, Department of Medicine, School of Medicine, East Carolina University, Greenville, NC, Sept. 1994 - 1998.

Senior Staff, National Institutes of Health, Bethesda, MD, 1990 - 1994.

Research Associate, Experimental Hematology, Armed Forces Radiobiology Research Institute, National Naval Medical Center, Bethesda, MD, 1987 - 1990.

Teaching Fellow, Boston University, 1984 - 1987.

Instructor, Northeastern University College, Boston, MA, 1980 - 1982.

Chemistry Teacher, Phitilas School, Patras, 1976 - 1977.

Research Assistant, Department of Biology, University of Patras, 1976 - 1977.

PROFESSIONAL SOCIETIES

- International Society for Experimental Hematology.
- American Society of Hematology.
- International Society for Hematotherapy & Graft Engineering.
- American Association for the Advancement of Science.
- American Association of Blood Banks.
- American Association of Clinical Chemistry.

PROFESSIONAL SERVICE

Reviewer for: Blood; Experimental Hematology; International Journal of Cell Cloning; Clinical Immunology and Immunopathology; NIDR Intramural Research Programs; FASEB; Nutrition and Cancer, an International Journal; Biochemical and Biophysical Research Communications; Cancer Therapy; British Journal of Cancer.

Director, Stem Cell Processing/Gene Therapy Lab, East Carolina University School of Medicine, Department of Medicine, Hematology/Oncology Section, Sept. 1994 - Present.

Co-Organizer for East Carolina University School of Medicine Cord Blood Banking Center, 1996-Present.

Member of the Graduate Faculty, East Carolina University. 1997-Present.

FAHCT Inspector for Stem Cell Processing Units, Foundation for Accreditation of Hemopoietic Cell Transplantation. 2000-Present.

CERTIFICATES

1. The College of American Pathologists. 2001 Surveys and Educational Anatomic Pathology Programs Certificate.

2. Department of Health & Human Services, Health Care Financial Administration. CLIA Certificate to Perform General Immunology and Hematology Tests. CLIA ID#: 34D0966563. January 28, 2002 to January 27, 2004.

3. The College of American Pathologists. 2002 Surveys and Educational Anatomic Pathology Programs Certificate.

4. The College of American Pathologists. 2003 Surveys and Educational Anatomic Pathology Programs Certificate.

5. Department of Health & Human Services, Health Care Financial Administration. CLIA Certificate to Perform General Immunology and Hematology Tests. CLIA ID#: 34D0966563. January 28, 2004 to January 27, 2006.

6. The College of American Pathologists. 2004 Surveys and Educational Anatomic Pathology Programs Certificate.

PATENTS

1. U.S. Divisional Patent SN 08/842,700. "Method of Treating Endothelial Injury".

2. "Method of Treating Endothelial Injury Using Chemotherapeutic Drugs". U.S. Divisional Patent Application Serial No. 09/525,797.

3. "rHuEPO can prevent and/or repair vascular injury Induced by Chemotherapeutic Agents." U.S. Divisional Patent Application Serial No. 09/525,808.

4. "Erythropoietin Ameliorates Chemotherapy-Induced Toxicity In Vivo". U.S. Patent Application Serial No. 10/117,011.

GRANTS

"Blood Micro-culture Assay of Hormone-Dependent Proliferation in Hematologic Disorders", Consultant, NIH, SBIR, 1988, $50,000.

"Blood Progenitors Assay for Hematologic Disorders", Principal Investigator, NIH, SBIR, 1989-1991, $500,000.

"Antineoplastic Properties of a Natural Stable Thioallyl", Principal Investigator, East Carolina University, School of Medicine, 1996-1997, $25,000.

"Role of Angiogenesis in Tumor Growth", Principal Investigator, Leo Jenkins Cancer Center, 1996-1997, $5,000.

"Prevention of Breast Cancer by Chemopreventors", Principal Investigator, U.S. Women's Open Golf Tournament Foundation, 1996-1997, $2,500.

"ERYTHROPOIETIN PROTECTS NORMAL ENDOTHELIUM AND IN COMBINATION WITH CHEMOTHERAPY DRUGS INHIBITS VESSEL FORMATION", Principal Investigator, Robert Wood Johnson Foundation, 1999-2002, $644,138.

In Vitro Correlates of Immunotherapy and Pharmacotherapy for Allergy and Asthma", Co-Investigator, Genentech, Inc., 2002-2005, $100,000.

"PROTECTIVE EFFECTS OF ERYTHROPOIETIN (EPO) ON PULMONARY FUNCTION AND DRUG-INDUCED FIBROSIS OF THE LUNGS", Principal Investigator, Robert Wood Johnson Foundation, 2001-2004, $442,068.

COMMITTEES

- Gene Targeting and Transgenics Committee, NIH, Bethesda, MD, 1992-1994.
- Centralized Facilities Committee, NIH, Bethesda, MD, 1992-1994.
- Radiation Safety Committee, NIH, Bethesda, MD, 1992-1994.
- Clinical Trials Committee, ECU, Greenville, NC, 1994 - Present.
- Department of Medicine, Research Committee, ECU, Greenville, NC, 1995 - Present.
- Task Force for Development of Promotions and Tenure Guidelines, ECU, Greenville, NC, 1997 - present.
- Graduate Studies Committee, ECU, Greenville, NC, 1997 - 1998.
- Administrative Section Coordinating Committee, American Association of Blood Banks, Bethesda, MD. 1998 – Present
- Bone Marrow Transplantation Patient Acceptance Team Committee, Pitt County Memorial Hospital. 1999-Present.
- Bone Marrow Transplantation Team Committee, Pitt County Memorial Hospital. 1999-Present.
- Bone Marrow Transplantation Issues Committee, Pitt County Memorial Hospital. 1999-Present.
- Bone Marrow Transplantation Quality Control Committee, Pitt County Memorial Hospital. 1999-Present.
- Eastern North Carolina Regional Center for Genomics and Bioinformatics Committee, East Carolina University. 2000-present.
- Genomics Planning Subcommittee, East Carolina University. 2001-present.
- Johnson and Johnson International Scientific Board. 2002.
- Education Subcommittee, L. J. Cancer Center. 2004 – present.

ADMINISTRATIVE ACTIVITIES

1. Responsible for the development and daily supervision of the Stem Cell Processing and Bioengineering Unit.

2. Responsible for compliance with the:
   - Foundation for the Accreditation of Cellular Therapies regulations
   - CLIA regulations
   - North American Task Force for Stem Cell Transplantation regulations
   - FDA regulations - Good Manufacturing Practices
   - OSHA regulations
   - Radiation Safety regulations

- Infectious Diseases regulations

3. Responsible for the writing and periodic editing of the Standard Operating Procedure Manual for the Clinical Stem Cell Processing Lab.

4. Management of daily operations of the Hematology/ Oncology Research Labs.

5. Supervise lab personnel on a daily basis.

6. Initiate work performance plans for each lab employee.

8. Prepare Radiation Safety protocols for radioisotope use.

9. Prepare protocols for animal use studies.

10. Prepare IRB protocols for human studies.

11. Participate in the selection and hiring of Laboratory Faculty and personnel.

12. Participate in the development of the Immunotherapy Program.

13. Participate in the overall development of the Bone Marrow Transplantation Program.

14. Prepare job descriptions and advertisements for positions in the Stem Cell Processing and Bioengineering Lab.

TEACHING ACTIVITIES

1976 - 1977	Pharmaceutical Chemistry; Organic Chemistry; Plant Physiology - Undergraduate and College students, University of Patras, Greece.
1980 - 1982	Histology Labs - Undergraduate students, Northeastern University.
1983 - 1984	Cell Biology - Undergraduate students, Boston University.
1984 - 1987	Embryogenesis; Morphogenesis; Comparative Anatomy - Undergraduate and Graduate students, Boston University.
1986 - 1987	Gross Anatomy Labs - Undergraduate students, Boston University.
1991 - 1994	Recombinant DNA; Immunology (Molecular and Conventional); Virology - Undergraduate, Graduate, and post Doctoral students, National Institute of Health.
1994-present	Tissue Culture Techniques; Recombinant DNA Technology; Molecular Immunology - Undergraduate and Graduate students, and Technicians, East Carolina University, School of Medicine.
1995-present	Organize the "Stem Cell Journal Club" Seminars.
1995-present	Organize the "Hematology/Oncology Translational Research" meeting/Seminars.
1995-present	Participate in teaching of the following courses: BIOL 6880, BIOL 4504, BIOL 5995, BIOL 6504.

1995-1998	Thesis Director to Graduate Student (Jason Ciaramella). Thesis title: Isolation, Characterization and Functional Activity of Human Placental Stromal Cells in Hematopoiesis (presented and approved May 1998).
1996- present	Educational Seminars in Basic and Clinical Departments.
1996-1999	Thesis Director to Graduate Student (Jean Hatfield). Thesis title: The Role of MRHF Cells in Supporting Hematopoiesis.
1996-1999	Thesis Director to Graduate Student (Dianne Harbour-Beal). Thesis title: Cell Cycling of Hematopoietic Stem Cells Cultured in FLT3-L and Thrombopoietin.
1996-present	Advise and direct undergraduate students in Research Projects and Courses.
2000-2003	Teaching Fellows "Immunobiology."
2001-2003	Teaching Residents "Principals in Biomedical Research and Technology."
2002-2003	Course Director: "Genetic Engineering and Transgenic Technology", a 6000 level graduate course.
2000-present	Advising Residents and Fellows in Biomedical Research.
2005-present	Thesis Committee Member for Graduate student Dave C. Francisco. Thesis Title: "Repair of oxidative clustered DNA lesions in breast cancer cells."
2003-present	Course Director: SCTH/BIOL 7210 SCTH/BIOL 7211 SCTH/BIOL 7212 SCTH/BIOL 7213

EDUCATIONAL MEETINGS AND TRAINING PROGRAMS ATTENDED

1. American Society of Hematology Meeting, Nashville, TN, December 1994.
2. "The Latest Advances and Strategies for Hematopoietic Stem cell Therapies" Symposium, Nashville, TN, December 1994.
3. "Engineering the Hematopoietic System" Symposium, Nashville, TN, December 1994.
4. Scholarly & Professional Writing Workshop, East Carolina University, Greenville, NC, February 1995.
5. Stem Cell Processing, Lab training session, NIH, Bethesda, MD, February 1995.
6. Stem Cell Processing, Lab training session, Georgetown, Washington, D.C., February 1995.
7. Clinical and Societal Issues in Blood & Marrow Transplantation for Hematologic Disease, Washington, DC, March 1995.
8. Stem Cell isolation - ex vivo expansion - cryopreservation - etc., Lab training session, NIH, Bethesda, MD, April 1995.

9. The International Society for Hematotherapy and Graft Engineering Meeting, Vancouver, Canada, June 1995.

10. Tumor Purging, Technical Workshop, Vancouver, Canada, June 1995.

11. Colony & LTCIC Assays & Culture Purging, Technical Workshop, Vancouver, Canada, June 1995.

12. Gene Transfer, Technical Workshop, Vancouver, Canada, June 1995.

13. Adoptive Immunotherapy, Technical Workshop, Vancouver, Canada, June 1995.

14. Stem Cell Collection, Technical Workshop, Vancouver, Canada, June 1995.

15. Cell Freezing, Storage & Infusion, Technical Workshop Vancouver, Canada, June 1995.

16. Fetal Tissue Transplantation, Workshop, San Francisco, CA, October 1995.

17. The Storage and Cryopreservation of Cord Blood, Workshop, San Francisco, CA, October 1995.

18. Second International Cord Blood Stem Cells, Conference, San Francisco, CA, October 1995.

19. Organizational Structure and Design of Stem Cell Processing Laboratory, NIH, Bethesda, MD, June 1996.

20. Stem Cell Processing and Bioengineering Laboratory Requirements, Georgetown, Washington, DC, June 1996.

21. Issues Related to Clinical Stem Cell Laboratory, University of Maryland, June 1996.

22. International Society for Experimental Hematology 1996, New York City, NY, August 1996.

23. American Society of Hematology Meeting, Orlando, FL, December 1996.

24. Safety of the Nation's Blood Supply, Orlando, FL, December 1996.

25. A Decade of Hematopoietic Growth Factors: Frontiers in Translational Research, Orlando, FL, December 1996.

26. Bone Marrow and Blood Cell Transplantation - Towards Component Therapy, Orlando, FL, December 1996.

27. Hematopoietic Inhibitors: Biology, Role in Cytosuppression, and Pharmacologic Modulation, Orlando, FL, December 1996.

28. Quality Control and Quality Assurance in the Stem Cell Transplantation Laboratory, Columbia, SC, December 1996.

29. North Carolina Cancer Research Symposium, Durham, NC, February 1997.

30. Peripheral Blood Stem Cells '97, International Society for Hematotherapy & Graft Engineering, Tempe, AZ, May 1997.

31. Peripheral Blood Stem Cell Collection, Interactive/ Practical Workshop - Baxter, Tempe, AZ, May 1997.

32. Peripheral Blood Stem Cell Collection, Interactive/ Practical Workshop - COBE-BCT, Tempe, AZ, May 1997.

33. Concentration and Cryopreservation, Interactive/Practical Workshop, Tempe, AZ, May 1997.

34. CD34 Selection, Interactive/Practical Workshop, Tempe, AZ, May 1997.

35. Cell Selection and Expansion, Interactive/Practical Workshop, Tempe, AZ, May 1997.

36. Purging and Cytokine Treatment, Interactive/Practical Workshop, Tempe, AZ, May 1997.

37. Stem Cell Processing and Cryopreservation, Columbia, SC, September 1997.

38. American Society of Hematology, San Diego, CA, December 1997.

39. New Advances in the use of Radioimmunotherapy for the Treatment of B Cell Lymphomas, San Diego, CA, December 1997.

40. Cell Selection: Broad Applications and Clinical Results, San Diego, CA, December 1997.

41. Monoclonal Antibody Therapies for Non-Hodgkins Lymphoma, San Diego, CA, December 1997.

42. Umbilical Cord Blood Bank, Stem Cell Collection and Processing Training, New York, NY, February 1998.

43. American Society of Hematology, Miami Beach, FL, December 1998.

44. Emerging Therapies for Hematologic Malignancies: Antibodies, Antisense Cytokines, and Anti-Inflammatory Approaches, Miami Beach, FL, December 1998.

45. Controversies in Stem Cell Transplantation, Miami Beach, FL, December 1998.

46. Sixth Annual Cell Selection Symposium: Engineered Cell Therapies, Miami Beach, FL, December 1998.

47. American Society of Hematology, New Orleans, LA, December 1999.

48. Immune-Based Therapies: The Coming of Age. New Orleans, LA, December 1999.

49. Novel Cytokines and Promising Strategies: From Cancer to Autoimmune Diseases. New Orleans, LA, December 1999.

50. New Advances in Biology and Treatment. New Orleans, LA, December 1999.

51. Foundation for Accreditation of Hemopoietic Cell Therapy Inspector Workshop, San Diego, CA, June 2000.

52. International Society for Hematotherapy and Graft Engineering, San Diego, CA, June 2000.

53. International Symposium for Molecular Hematology, New York, NY, July 2000.

54. ISHAGE Good Manufacturing Practices (GMP) 2000 Workshop, San Francisco, CA, December 2000.

55. Immune Mediators in the Genesis and Treatment of Cancer, San Francisco, CA, December 2000.

56. Cord Blood Banking and Transplantation Update, San Francisco, CA, December 2000.

57. Scientific and Technological Innovations in Biology, San Francisco, CA, December 2000.

58. Expanding the Promise of Stem-Cell Transplantation, San Francisco, CA, December 2000.

59. New Strategies and Modalities for Optimal Patient Outcomes, San Francisco, CA, December 2000.

60. American Society of Hematology, San Francisco, CA, December 2000.

61. FAHCT Inspector Continuing Education Workshop, Quebec City, Quebec, Canada, June 2001.

62. The International Society for Hematotherapy and Graft Engineering Annual Meeting, Quebec City, Quebec, Canada, June 2001.

63. Legal and Regulatory Affairs Workshop, Quebec City, Quebec, Canada, June 2001.

64. Gene Therapy Workshop, Quebec City, Quebec, Canada, June 2001.

65. Graft Evaluation Workshop, Quebec City, Quebec, Canada, June 2001.

66. Immunotherapy Workshop, Quebec City, Quebec, Canada, June 2001.

67. Hematopoietic Stem Cell Gene Therapy: From Novel Technologies to Clinical Trials, Tokyo, Japan, August 2001.

68. Clinical and Immunological Correlates of Stem Cell Transplantation in Rheumatic Autoimmune Disease, Tokyo, Japan, August 2001.

69. Annual Meeting of the International Society for Experimental Hematology, Tokyo, Japan, August 2001.

70. Emerging Cellular Therapies: Enhancing the Human Hematopoietic and Immune Systems, Orlando, FL, December 2001.

71. Large Scale Separation Enabling Novel Treatment Strategies for Cellular Therapy, Orlando, FL, December 2001.

72. The Promise of Cellular Therapy, Orlando, FL, December 2001.

73. The 43rd Annual Meeting of the American Society of Hematology, Orlando, FL, December, 2001.

74. Hypothesis Generating Workshop. Potential Impact of rHuEPO Treatment on Prognosis ,

Mode of Actions, Clinical Evidence. New York, NY, May 2002.

75. University and Medical Center Institutional Review Board , East Carolina University. Certificate of training. September 2002.

76. The College of American Pathologists. Surveys and Educational Anatomic Pathology Programs. Certificate of Participation for 2002.

77. Bringing Ideas to Market Workshop, East Carolina University. March 2003.

78. The 9th Annual Meeting of the International Society of Cell Therapies, Phoenix, AZ, May 2003.

79. The Potential of Cellular Engineering and Therapy Symposium, Phoenix, AZ, May 2003.

80. Symposium on Emerging Strategies Utilizing Cellular Therapies to Rebuild Tissue and Immune Function in Patients, Phoenix, AZ, May 2003.

81. Analysis of Immunoreconstitution Post-Transplant (Spectratyping, TREC Assays, Eli Spots) Technical Workshop, Phoenix, AZ, May 2003.

82. Immunotherapy Workshop, Phoenix, AZ, May 2003.

83. Evaluation of the Apheresis Product – from Collection to Reinfusion Technical Workshop, Phoenix, AZ, May 2003.

84. Non-Hematopoietic and Mesenchymal Stem Cells Workshop, Phoenix, AZ, May 2003.

85. Dendritic Cell Preparation Technical Workshop, Phoenix, AZ, May 2003.

86. The 32nd Annual Meeting of the International Society for Experimental Hematology, Paris, France, July 2003.

87. Emergent Therapies Symposium, Paris, France, July 2003.

88. State of the Art in Signal Transduction Inhibition in Hematology Symposium, Paris, France, 2003.

89. Cell-Dyn 3700 Hematology Training, Abbott Diagnostics Division, Irving, TX, November 2003.

90. Symposium on Cord Blood Transplantation: From Science to Practical Applications, San Diego, CA, December 2004.

91. Symposium on Cellular Therapy – Progressing to clinical Practice, San Diego, CA, December 2004.

92. New Clinical Strategies and Emerging Research in Multiple Myeloma Symposium, San Diego, CA, December 2004.

93. The 46th Annual Meeting of the American Society of Hematology, San Diego, CA, December 2004.

RESEARCH ACTIVITIES AND INTERESTS

1. A long-term goal is to detect and characterize natural or pharmacological inhibitors and stimulators of angiogenesis, and apply them in the treatment of patients with various cancers.

2. A translational research project of immense interest to our cancer program is the isolation, characterization, and EX VIVO expansion of lymphohematopoietic and mesenchymal stem cells.

3. My research activities include studies on micronutrients (e.g. vitamin E and organosulfurs, etc.) in cancer prevention and treatment.

PUBLICATIONS

Articles (Refereed):

Monette, F.C. and **Sigounas, G.**: Factors affecting the proliferation and differentiation of clonogenic hematopoietic stem cells in vitro. Blood Cells 10:261-274, 1984.

Monette, F.C. and **Sigounas, G.**: The sensitivity of murine multipotential stem cell colony (CFU-GEMM) growth in interleukin-3, erythropoietin and hemin. Exp. Hematol. 15:729-734, 1987.

Hankins, W.D., Chin K., and **Sigounas, G.**: Hormone associated therapy of leukemia: reflections. *In* Hormone, Cell Biology and Cancer. Prospectives and Potential. Alan R. Liss, New York. Prog. Clin. Biol. Res. 262:257-267, 1988.

Monette, F.C. and **Sigounas, G.**: Growth of murine multipotent stem cells in a simple "serum-free" culture system: Role of interleukin-3, erythropoietin, and hemin. Exp. Hematol. 16:250-255, 1988.

Monette, F.C. and **Sigounas, G.**: Hemin acts synergistically with IL-3 to promote the growth of multipotent stem cells (CFU-GEMM) in serum-free cultures of normal murine bone marrow. Exp. Hematol. 16:727-729, 1988.

Hankins, W.D., Chin, K., and **Sigounas, G.**: Erythropoietin-dependent and erythropoietin-producing erythroid cell lines: Practical and therapeutic implications. Annals NY Acad. Sci. 554, 21-28, 1989.

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Abstracts:

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Niskanen, E., Williams, M.E., and **Sigounas, G.**: Repopulation of lethally irradiated mice by cells derived from colonies formed in diffusion chambers in mice. Blood 75 (Suppl. 1):109a, 1990.

Sigounas, G., MacVittie, T.J., and Jaenisch, R.: Transgenic mice: A model system to study lymphohematopoietic growth factors in vivo. J. Cell. Biochem (Suppl. 1) 14E:99, 1990.

Sigounas, G., Jaenisch, R., and MacVittie, T.J.: Transgenic hematopoietic stem cells (HSC) as probes for lympho-hematopoiesis and experimental transplantation. Exp. Hematol. 18:704, 1990.

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Steiner, M., Ciaramella, J., Rollins, L., Anagnostou, A., and **Sigounas, G.**: Inhibition of protein kinase C activity by α-tocopherol blocks PMA induced shape change of HEL cells. East Carolina University, School of Medicine, Department of Medicine, Tenth Annual Research Day: March 7, 1996.

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Rollins, L., Madigan, T.C., Steiner, M., **Sigounas, G.**, Anagnostou, A.A.: The cloning and analysis of the erythropoietin receptor from human endothelial cells. East Carolina University, School of Medicine, Department of Medicine, Tenth Annual Research Day: March 7, 1996.

Sigounas, G., Hooker, J., Anagnostou, A., Steiner, M.: S-Allylmercaptocysteine inhibits cell proliferation and reduces the viability of breast and prostate cancer cell lines. 1997 North Carolina Cancer Research Symposium: February 1997.

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Madigan, T., Anagnostou, A., Steiner, M., **Sigounas, G.**: Cloning and Characterization of the erythropoietin receptor of vascular endothelial cells. East Carolina University, School of Medicine, Department of Medicine, Eleventh Annual Research Day: March 6, 1997.

Ciaramella, J., Steiner, M., Anagnostou, A., **Sigounas, G.**: A human fibroblast cell line can substitute for marrow stromal cells and promote the proliferation and development of multipotent hematopoietic stem cells. East Carolina University, School of Medicine, Department of Medicine, Eleventh Annual Research Day: March 6, 1997.

Hooker, J., Steiner, M., Anagnostou, A., **Sigounas, G.**: Interaction of erythropoietin with chemotherapeutic agents. Potential therapy for highly vascularized tumors. East Carolina University, School of Medicine, Department of Medicine, Eleventh Annual Research Day: March 6, 1997.

Pei, X., Ciaramella, J., Steiner, M., Anagnostou, A., **Sigounas, G.**: Immortalization of human placental stromal cells. East Carolina University, School of Medicine, Department of Medicine, Eleventh Annual Research Day: March 6, 1997.

Pei, X., Hooker, S., Anagnostou, A., **Sigounas, G.**, Steiner, M.: S-Allymercaptocysteine (SAMC) changes regulation of cell cycle. East Carolina University, School of Medicine, Department of Medicine, Eleventh Annual Research Day: March 6, 1997.

Anagnostou, A., Madigan, T., Steiner, M., **Sigounas, G.**: Erythropoietin Receptor of Human Vascular Endothelial Cells: Cloning, Characterization and Putative Function. Acta Haematologica, (Suppl. A) 98:32, 1997.

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Sigounas, G., Hooker, J., and Anagnostou, A.: The Role of EPO in Neovascularization: Synergistic Anti-tumor and Anti-angiogenic Effect of EPO and Chemotherapeutic Drugs. East Carolina University, School of Medicine, Department of Medicine Twelfth Annual Research Day, March 5, 1998.

Hooker, J., Steiner, M., Anagnostou, A., and **Sigounas, G.**: The Role of S-Allylmercap-tocysteine (SAMC) on Expression and Function of v-HA-RAS Gene. East Carolina University, School of Medicine, Department of Medicine Twelfth Annual Research Day, March 5, 1998.

Beal, D., Anagnostou, A, and **Sigounas, G.**: Cell Cycle Status of Human Umbilical Cord Blood-Derived CD34+Cells Treated with Thrombopoietin and FLT3-Ligand. East Carolina University, School of Medicine, Department of Medicine Twelfth Annual Research Day, March 5, 1998.

Harbour-Beal, D., Pei, XF., Anagnostou, A., **Sigounas, G.**: Cell Cycling of Flt3 Ligand/Thrombopoietin-Treated CD34+ Hemopoietic Stem Cells. East Carolina University, School of Medicine, Department of Medicine Thirteenth Annual Research Day, March 9, 1999.

Hatfield, J., Ciramella, J., Anagnostou, A., **Sigounas, G.**: The Human Firbroblast MRHF Cells are Capable of Supporting the *Ex Vivo* Expansion of CD34+ Cells. East Carolina University, School of Medicine, Department of Medicine Thirteenth Annual Research Day, March 9, 1999.

Sigounas, G., Manning, C., Powell, M., Comeau T., Schadler, L.: Peripheral Blood CD34+ Cell Counts and the Colony-forming Cells Can Predict the Effectiveness of Mobilization and Determine Both the Number of Stem Cell Collections and the Blood Volume Required for Leukapheresis. East Carolina University, School of Medicine, Department of Medicine Fourteenth Annual Research Day, March 16, 2000.

Mehlhop, P., Sigounas, A., **Sigounas, G.**: Enhancement of IL-4 and IFN-γ Production by Peripheral Blood Mononuclear Cells in Patients on Allergen Immunotherapy. East Carolina University, School of Medicine, Department of Medicine 15th Annual Research Day, May 24, 2001.

Sigounas, G., Mehlhop, P., Sigounas, A., Collins, M., Lee, W.: Erythropoietin Ameliorates Chemotherapy-Induced Toxicity In Vivo. Exp. Hematol. 29(8): 57, 2001.

Sigounas, G., Salleng, K., Mehlhop, P., Sallah, S., Collins, M., Kardatos, A., Sigounas, D.: Erythropoietin Inhibits Drug-Induced Fibrosis and Improves Pulmonary Function. Blood 98(11): 798a, 2001.

Howell, E., Mehlhop, P., Sigounas, A., **Sigounas, G.**, Patel, D.: IL-4 Production in Response to Allergen Stimulation and Clinical Symptoms of Allergic Rhinitis. American Academy of Allergy, Asthma and Immunology 58th Annual Meeting, March, 2002.

Sallah, S., Wan, J., **Sigounas, G.**, Nguyen, N.: Recombinant Activated Factor VII Can Control the Bleeding Manifestations of Disseminated Intravascular Coagulation in Patients with Cancer. Blood 100(11): 99b, 2002.

Sallah, S., Wan, J., Nguyen, N., **Sigounas, G.**: Combination Imatinib and Gemtuzumab Ozogamicin in Patients with Chronic Myeloid Leukemia in Myeloid Blast Crisis. Blood 100(11): 319b, 2002.

Sigounas, G., Collins, M., Sallah, S., Sigounas, V.: Erythropoietin Enhances Antineoplastic Activity of Chemotherapeutic Agents. Blood 100(11): 210, 2002.

Sigounas, G., Salleng, K., Collins, M., Sigounas, D.: Erythropoietin Inhibits Endothelial Cell Activation and Intercellular Adhesion Molecule-1 Expression. Exp. Hematol. 31(7,1): 79, 2003.

Sallah, A.S., Husain, A., Wan, J., Turturro, F., **Sigounas, G.**, Glass, J.: Oral anticoagulation in patients with solid tumors and deep venous thrombosis: Bleeding and clotting events and monitoring of plasma coagulation markers. Blood 102 (11): 323a, 2003.

Sallah, A.S., Husain, A., Wan, J., **Sigounas, G.**: Markers of activation of hemostasis in patients with solid tumors: Correlation with progression and prognosis. Blood 102 (11): 89b, 2003.

Brick, W.G., Quan, W.D., Shah, R., Khan, N., Sigounas, G., Burgess, R.E.: Treatment of a bilaterally nephrectomizes patient with recombinant IFNα-2b (IFN): Pharmacokinetic evaluation suggests that approximately 50% of serum IFN is eliminated by hemodialysis. The 95th Annual Meeting of American Association for Cancer Research. Proceedings of the AACR, p. 270, 2004.

PRESENTATIONS/Invited Speaker

"Transgenic Transplantation: A New in Vivo System for Experimental Hemopoiesis which Employs Sequential Molecular Monitoring of Multiple Genetic Markers." Plenary Session, at the 31st Annual Meeting of American Society of Hematology, Atlanta, Georgia, December 3, 1989.

"Ex vivo and in Vivo Analysis of Transgenic Hematopoietic Stem Cells." USHUS, Bethesda, Maryland, February 20, 1990.

"Transgenic Mice: A Model System to Study Lymphohematopoietic Growth Factors in Vivo." At the 19th Annual Meeting of UCLA Symposia on Molecular & Cellular Biology, Steamboat Springs, Colorado, April 3, 1990.

"Transgenic Hematopoietic Stem Cells (HSC) as Probes for Lymphohematopoiesis and Experimental Transplantation." Presidential Symposium, at the 19th Annual Meeting of International Society for Experimental Hematology, Seattle, Washington, August 30, 1990.

"Treatment of Human Genetic Disorders: Natural vs Genetically Engineered Approaches." NIH, Bethesda, Maryland, January 11, 1991.

"Establishment of the Human Immune System in SCID Mice." NIH, Bethesda, Maryland, March 5, 1991.

"Biological Properties of Polyreactive Antibodies." NIH, Bethesda, Maryland, March 3, 1992.

"Cloning, Structure and Function of the CD5 Gene." NIH, Bethesda, Maryland, November 3, 1992.

"Cell Type that Makes Polyreactive Antibodies." NIH, Bethesda, Maryland, November 10, 1992.

"Physiological and Pathological Functions of Polyreactive Antibodies." NIH, Bethesda, Maryland, November 17, 1992.

"Role of Apoptosis in Immunoregulation and Removal of Autoreactive T cells." NIH, Bethesda, Maryland, January 24, 1993.

"Ebstein Barr Virus (EBV) Receptors" NIH, Bethesda, Maryland, March 5, 1993.

"DNA Vaccines."NIH, Bethesda, Maryland, February 18, 1994.

"Targeting the CD5 Gene." NIH, Bethesda, Maryland, April 5, 1994.

"The Role of Erythropoietin in Neovascularization." ECU School of Medicine, Ninth Annual Department of Medicine Research Day, June 1, 1995.

"Anticancer Activity of Organosulfurs." ECU School of Medicine, Department of Microbiology and Immunology, January 31, 1996.

"The Transgenic Mouse System." ECU School of Medicine, Department of Physiology, February 8, 1996.

"Antineoplastic Properties of S-Allylmercaptocysteine." East Carolina University, School of Medicine, Tenth Annual Department of Medicine Research Day, March 7, 1996.

"Alpha-Tocopherol Induces Apoptosis and Changes of p53 Expression in Erythroleukemia Cells. Experimental Biology 96, Washington, D.C., April 15, 1996.

"Inhibition of Protein Kinase C Activity by α-Tocopherol (α-T) Blocks PMA-Induced Shape Change of HEL Cells. Experimental Biology 96, Washington, D.C., April 17, 1996.

"The Role of Biofactors Micronutrients in Cancer Prevention" East Carolina University, School of Medicine, Department of Medicine, Hematology/Oncology Evening Grand Rounds, April 25, 1996.

"Stromal Cell Lines That Support Stem Cell Growth" East Carolina University, School of Medicine, Department of Medicine, Hematology/Oncology Journal Club, January 21, 1997.

"S-Allylmercaptocysteine Inhibits Cell Proliferation and Reduces the Viability of Breast and Prostate Cancer Cell Lines", 1997 North Carolina Cancer Research Symposium, Durham, NC, February 7, 1997.

"S-Allylmercaptocysteine Induces Apoptosis in Erythroleukemia Cell Lines" 1997 North Carolina Cancer Research Symposium, Durham, NC, February 7, 1997.

"Cloning and Characterization of the Erythropoietin Receptor of Vascular Endothelial Cells" East Carolina University, School of Medicine, Department of Medicine Eleventh Annual Research Day, March 6, 1997.

"A Human Fibroblast Cell Line Can Substitute for Marrow Stromal Cells and Promote the Proliferation and Development of Multipotent Hematopoietic Stem Cells" East Carolina University, School of Medicine, Department of Medicine Eleventh Annual Research Day, March 6, 1997.

"Interaction of Erythropoietin with Chemotherapeutic Agents. Potential Therapy for Highly Vascularized Tumors" East Carolina, University School of Medicine, Department of Medicine Eleventh Annual Research Day, March 6, 1997.

"S-Allylmercaptocysteine (SAMC) Changes Regulation of Cell Cycle" East Carolina University, School of Medicine, Department of Medicine Eleventh Annual Research Day, March 6, 1997.

"Immortalization of Human Placental Stromal Cells" East Carolina University, School of Medicine, Department of Medicine Eleventh Annual Research Day, March 6, 1997.

"Erythropoietin and Epo Receptors in Non-Erythroid Cells" East Carolina University, School of Medicine, Immunology Journal Club, March 19, 1997.

"Gene Targeting and the Knockout Technology" East Carolina University, School of Medicine, Department of Physiology, April 15, 1997.

"Future Cancer Therapies: Can Erythropoietin Synergize with Cisplatin to Suppress Tumor Growth?", East Carolina University, School of Medicine, Department of Medicine, Hematology/Oncology Educational Conference, May 9, 1997.

"Breast Cancer Research within the Division of Hematology/Oncology." Breast Cancer Research Retreat, East Carolina University School of Medicine, Greenville, NC, November 24, 1997.

"DNA Manipulation for Gene Therapy and Transgenics", North Carolina Association for Biomedical Research, at East Carolina University School of Medicine, Greenville, NC, January 16, 1998.

"Anti-angiogenic Action of Anti-neoplastic Drugs; Biomodulation of this Activity by Recombinant EPO", East Carolina University School of Medicine, Department of Medicine, Hematology/Oncology Educational Conference, February 13, 1998.

"The Role of EPO in Neovascularization: Synergistic Anti-tumor and Anti-angiogenic Effect of EPO and Chemotherapeutic Drugs". East Carolina University, School of Medicine, Department of Medicine Twelfth Annual Research Day, March 5, 1998.

"The Role of S-Allylmercaptocysteine (SAMC) on Expression and Function of v-HA-RAS Gene". East Carolina University, School of Medicine, Department of Medicine Twelfth Annual Research Day, March 5, 1998.

"Cell Cycle Status of Human Umbilical Cord Blood-Derived CD34+Cells Treated with Thrombopoietin and FLT3-Ligand". East Carolina University, School of Medicine, Department of Medicine Twelfth Annual Research Day, March 5, 1998.

"Stem Cell Transplantation Therapies for Malignant and Non-Malignant Disorders". East Carolina University, School of Medicine, Microbiology & Immunology Journal Club, Greenville, NC, May 6, 1998.

"Present and Future Hematopoietic Stem Cell Therapies" East Carolina University School of Medicine, Department of Medicine, Section of Allergy Grand Rounds, Greenville, NC, May 7, 1998.

"The role of Epo and Chemotherapeutic Drugs in Neovascularization and Tumor Inhibition." University of Ioannina, Greece. July 1998.

"Stem Cell Transplantion for Malignant and Non-Malignant Disorders." University of Ioannina, Greece. July 1998.

"Synergistic Anti-angiogenic and anti-Tumor Effect of Erythropoietin and Chemotherapeutic Drugs", East Carolina University School of Medicine, Department of Microbiology and Immunology, Immunobiology Journal Club, Greenville, NC, February 17, 1999.

"Cell Cycling of Flt3 Ligand/Thrombopoietin-Treated CD34+ Hemopoietic Stem Cells." East Carolina University, School of Medicine, Department of Medicine Thirteenth Annual Research Day, March 9, 1999.

"The Human Fibroblast MRHF Cells are Capable of Supporting the *ExVivo* Expansion of CD34+ Cells." East Carolina University, School of Medicine, Department of Medicine Thirteenth Annual Research Day, March 9, 1999.

"Evolution and Applications of Gene Therapy." East Carolina University School of Medicine, Department of Physiology, April 15, 1999.

"Peripheral Blood CD34+ Cell Counts and the Colony-forming Cells Can Predict the Effectiveness of Mobilization and Determine Both the Number of Stem Cell Collections and the Blood Volume Required for Leukapheresis." East Carolina University, School of Medicine, Department of Medicine Fourteenth Annual Research Day, March 16, 2000.

"The Future of Stem Cell Therapies in Treating Malignant Disorders", East Carolina University School of Medicine, Department of Microbiology and Immunology, Immunobiology Journal Club, Greenville, NC, March 8, 2000.

"Advantages, Drawbacks, and Future Directions of Cellular Therapies in the Treatment of Malignant Disorders". Department of Medicine, Section of Allergy Grand Rounds, Greenville, NC, March 23, 2000.

"Erythropoietin Ameliorates Chemotherapy-Induced Toxicity In Vivo". 30th Annual Meeting of the International Society for Experimental Hematology, Tokyo, Japan, August 27, 2001.

"Erythropoietin Inhibits Drug-Induced Fibrosis and Improves Pulmonary Function". Forty-third Annual Meeting of the American Society of Hematology, Orlando, Florida, December 10, 2001.

"Pleotropic Properties of Erythropoietin." East Carolina University, Brody School of Medicine, Department of Microbiology and Immunology, Immunobiology Journal Club, Greenville, NC, February 19, 2003.

"IL-4 Production in Response to Allergen Stimulation and Clinical Symptoms of Allergic Rhinitis." East Carolina University, Brody School of Medicine, Department of Internal Medicine Seventeenth Annual Research Day, April 2003.

"Erythropoietin Inhibits Endothelial Cell Activation and Intercellular Adhesion Molecule-1 Expression." 32nd Annual Meeting of the International Society for Experimental Hematology, Paris, France, July 6, 2003.

"Pleiotropic Actions of Erythropoietin" Department of Hematology and the Hematology Laboratory, Ioannina University Medical School/University Hospital, Greece. February 17, 2004.

"Hematopoietic Stem Cell Present and Future Therapies." Department of Biology, East Carolina University. November 4, 2004.

"Stromal and Hematopoietic Stem Cell Properties and Therapies." Diabetes Group. Brody School of Medicine. March 28, 2005.

AWARDS AND OTHER HONORS

1972 & 1976	National Institute Scholarships, Greece.
1984-1987	Teaching Fellowship, Boston University. Graduate School Scholarship, Boston University. Research Assistantship, Boston University.
1986	International Society for Experimental Hematology (ISEH) Travel Award.
1987-1990	Research Associateship, National Academy of Science, Washington, DC.
1998	"Best Abstract Award", Department of Medicine Twelfth Annual Research Day, East Carolina University School of Medicine, Greenville, NC

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...administration, parenteral drug products should ... ected visually for particulate matter and discolor... Because ORTHOCLONE OKT3 is a protein solu... may develop a few fine translucent particles ... have been shown not to affect its potency.

...teriostatic agent is present in this product; adher... aseptic technique is advised. Once the ampule is ... use immediately and discard the unused portion. ... ORTHOCLONE OKT3 for injection by drawing ... into a syringe through a low protein-binding 0.2 micrometer (μm) filter. Discard filter and attach a ...dle for intravenous bolus injection.

...no data is available on compatibility of ...OCLONE OKT3 with other intravenous sub... or additives, other medications/substances ...not be added or infused simultaneously through ... intravenous line. If the same intravenous line is ... for sequential infusion of several different drugs, ... should be flushed with saline before and after ...on of ORTHOCLONE OKT3.

...ister ORTHOCLONE OKT3 as an intravenous ... less than one minute. Do **not** administer by intra... infusion or in conjunction with other drug ...ions.

...SUPPLIED

...LONE OKT3 is supplied as a sterile solution in ... 5 ampules (NDC 59676-101-01). Each 5 mL ...ntains 5 mg of muromonab-CD3.

...Store in a refrigerator at 2° to 8°C (36° to 46°F). DO ...FREEZE OR SHAKE.

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...in is a glycoprotein which stimulates red blood ... It is produced in the kidney and stimulates ... differentiation of committed erythroid pro... the bone marrow. PROCRIT (Epoetin alfa), a 165 ... glycoprotein manufactured by recombinant DNA ... has the same biological effects as endogenous ... It has a molecular weight of 30,400 daltons ... by mammalian cells into which the human erythropoietin gene has been introduced. The product contains the identical amino acid sequence of isolated natural erythropoietin.

PROCRIT is formulated as a sterile, colorless, liquid in an isotonic sodium chloride/sodium citrate buffered solution for intravenous (IV) or subcutaneous (SC) administration.

Single-Dose, Preservative-Free Vial: Each 1 mL of solution contains 2,000, 3,000, 4,000 or 10,000 units of Epoetin alfa, 2.5 mg Albumin (Human), 5.8 mg sodium citrate, 5.8 mg sodium chloride, and 0.06 mg citric acid in Water for Injection, USP (pH 6.9±0.3). This formulation contains no preservative.

Multidose, Preserved Vial: 2 mL (20,000 units, 10,000 Units/mL). Each 1 mL of solution contains 10,000 units of Epoetin alfa, 2.5 mg Albumin (Human), 1.3 mg sodium citrate, 8.2 mg sodium chloride, 0.11 mg citric acid, and 1% benzyl alcohol as preservative in Water for Injection, USP (pH 6.1±0.3).

CLINICAL PHARMACOLOGY

Chronic Renal Failure Patients

Endogenous production of erythropoietin is normally regulated by the level of tissue oxygenation. Hypoxia and anemia generally increase the production of erythropoietin, which in turn stimulates erythropoiesis.[2] In normal subjects, plasma erythropoietin levels range from 0.01 to 0.03 Units/mL,[2,3] and increase up to 100- to 1000-fold during hypoxia or anemia.[2,3] In contrast, in patients with chronic renal failure (CRF), production of erythropoietin is impaired, and this erythropoietin deficiency is the primary cause of their anemia.[3,4]

Chronic renal failure is the clinical situation in which there is a progressive and usually irreversible decline in kidney function. Such patients may manifest the sequelae of renal dysfunction, including anemia, but do not necessarily require regular dialysis. Patients with end-stage renal disease (ESRD) are those patients with CRF who require regular dialysis or kidney transplantation for survival.

PROCRIT has been shown to stimulate erythropoiesis in anemic patients with CRF, including both patients on dialysis and those who do not require regular dialysis.[4–13] The first evidence of a response to the three times weekly (T.I.W.) administration of PROCRIT is an increase in the reticulocyte count within 10 days, followed by increases in the red cell count, hemoglobin, and hematocrit, usually within 2–6 weeks.[4,5] Because of the length of time required for erythropoiesis—several days for erythroid progenitors to mature and be released into the circulation—a clinically significant increase in hematocrit is usually not observed in less than 2 weeks and may require up to 6 weeks in some patients. Once the hematocrit reaches the suggested target range (30–36%), that level can be sustained by PROCRIT therapy in the absence of iron deficiency and concurrent illnesses.

The rate of hematocrit increase varies between patients and is dependent upon the dose of PROCRIT, within a therapeutic range of approximately 50-300 Units/kg (T.I.W.).[4] A greater biologic response is not observed at doses exceeding 300 Units/kg (T.I.W.).[6] Other factors affecting the rate and extent of response include availability of iron stores, the baseline hematocrit, and the presence of concurrent medical problems.

Zidovudine-treated HIV-infected Patients

Responsiveness to PROCRIT in HIV-infected patients is dependent upon the endogenous serum erythropoietin level prior to treatment. Patients with endogenous serum erythropoietin levels ≤500 mUnits/mL, and who are receiving a dose of zidovudine ≤4,200 mg/week, may respond to PROCRIT therapy. Patients with endogenous serum erythropoietin levels >500 mUnits/mL do not appear to respond to PROCRIT therapy. In a series of four clinical trials involving 255 patients, 60% to 80% of HIV-infected patients treated with zidovudine had endogenous serum erythropoietin levels ≤500 mUnits/mL.

Response to PROCRIT in zidovudine-treated HIV-infected patients is manifested by reduced transfusion requirements and increased hematocrit.

Cancer Patients on Chemotherapy

Anemia in cancer patients may be related to the disease itself or the effect of concomitantly administered chemotherapeutic agents. PROCRIT has been shown to increase hematocrit and decrease transfusion requirements after the first month of therapy (months 2 and 3), in anemic cancer patients undergoing chemotherapy.

A series of clinical trials enrolled 131 anemic cancer patients who were receiving cyclic cisplatin- or non cisplatin-containing chemotherapy. Endogenous baseline serum erythropoietin levels varied among patients in these trials with approximately 75% (N=83/110) having endogenous serum erythropoietin levels ≤132 mUnits/mL, and approximately 4% (N=4/110) of patients having endogenous serum erythropoietin levels >500 mUnits/mL. In general, patients with lower baseline serum erythropoietin levels responded more vigorously to PROCRIT than patients with higher baseline erythropoietin levels. Although no specific serum erythropoietin level can be stipulated above which patients would be unlikely to respond to PROCRIT therapy, treatment of patients with grossly elevated serum erythropoietin levels (e.g., >200 mUnits/mL) is not recommended.

Pharmacokinetics

Intravenously administered PROCRIT is eliminated at a rate consistent with first order kinetics with a circulating half-life ranging from approximately 4 to 13 hours in patients with CRF. Within the therapeutic dose range, detectable levels of plasma erythropoietin are maintained for at least 24 hours.[7] After subcutaneous administration of PROCRIT to patients with CRF, peak serum levels are achieved within 5–24 hours after administration and decline slowly thereafter. There is no apparent difference in half-life between patients not on dialysis whose serum creatinine levels were greater than 3, and patients maintained on dialysis.

In normal volunteers, the half-life of intravenously administered PROCRIT is approximately 20% shorter than the half-life in CRF patients. The pharmacokinetics of PROCRIT have not been studied in HIV-infected patients.

INDICATIONS AND USAGE

Treatment of Anemia of Chronic Renal Failure Patients

PROCRIT is indicated in the treatment of anemia associated with chronic renal failure, including patients on dialysis (end-stage renal disease) and patients not on dialysis. PROCRIT is indicated to elevate or maintain the red blood cell level (as manifested by the hematocrit or hemoglobin determinations) and to decrease the need for transfusions in these patients.

PROCRIT is not intended for patients who require immediate correction of severe anemia. PROCRIT may obviate the need for maintenance transfusions but is not a substitute for emergency transfusion.

Prior to initiation of therapy, the patient's iron stores, including transferrin saturation and serum ferritin, should be evaluated. Transferrin saturation should be at least 20% and ferritin at least 100 ng/mL. Blood pressure should be adequately controlled prior to initiation of PROCRIT therapy, and must be closely monitored and controlled during therapy. Non-dialysis patients with symptomatic anemia considered for therapy should have a hematocrit less than 30%. All patients on PROCRIT therapy should be regularly monitored (see "PRECAUTIONS").

PROCRIT should be administered under the guidance of a qualified physician (see "Dosage and Administration").

Treatment of Anemia in Zidovudine-treated HIV-Infected Patients

PROCRIT is indicated for the treatment of anemia related to therapy with zidovudine in HIV-infected patients. PROCRIT is indicated to elevate or maintain the red blood cell level (as manifested by the hematocrit or hemoglobin determinations) and to decrease the need for transfusions in these patients. PROCRIT is not indicated for the treatment of anemia in HIV-infected patients due to other factors such as iron or folate deficiencies, hemolysis or gastrointestinal bleeding, which should be managed appropriately.

PROCRIT, at a dose of 100 Units/kg three times per week, is effective in decreasing the transfusion requirement and increasing the red blood cell level of anemic, HIV-infected patients treated with zidovudine, when the endogenous serum erythropoietin level is ≤500 mUnits/mL and when patients are receiving a dose of zidovudine ≤4,200 mg/week.

Treatment of Anemia in Cancer Patients on Chemotherapy

PROCRIT is indicated for the treatment of anemia in patients with non-myeloid malignancies where anemia is due to the effect of concomitantly administered chemotherapy. PROCRIT is indicated to decrease the need for transfusions in patients who will be receiving concomitant chemotherapy for a minimum of 2 months. PROCRIT is not indicated for the treatment of anemia in cancer patients due to other factors such as iron or folate deficiencies, hemolysis or gastrointestinal bleeding which should be managed appropriately.

Clinical Experience: Response to PROCRIT

Chronic Renal Failure Patients

Response to PROCRIT was consistent across all studies. In the presence of adequate iron stores (see "Pre-Therapy Iron Evaluation"), the time to reach the target hematocrit is a function of the baseline hematocrit and the rate of hematocrit rise.

The rate of increase in hematocrit is dependent upon the dose of PROCRIT administered and individual patient variation. In clinical trials at starting doses of 50–150 Units/kg (T.I.W.), patients responded with an average rate of hematocrit rise of:

HEMATOCRIT INCREASE

STARTING DOSE (T.I.W. IV)	POINTS/DAY	POINTS/2 WEEKS
50 Units/kg	0.11	1.5
100 Units/kg	0.18	2.5
150 Units/kg	0.25	3.5

Continued on next page

Ortho Biotech—Cont.

Over this dose range, approximately 95% of all patients responded with a clinically significant increase in hematocrit, and by the end of approximately 2 months of therapy virtually all patients were transfusion-independent. Changes in the quality of life of patients treated with PROCRIT were assessed as part of a Phase III clinical trial.[5,8] Once the target hematocrit (32-38%) was achieved, statistically significant improvements were demonstrated for most quality of life parameters measured, including energy and activity level, functional ability, sleep and eating behavior, health status, satisfaction with health, sex life, well-being, psychological effect, life satisfaction, and happiness. Patients also reported improvement in their disease symptoms. They showed a statistically significant increase in exercise capacity (VO_2 max), energy, and strength with a significant reduction in aching, dizziness, anxiety, shortness of breath, muscle weakness, and leg cramps.[8,14]

Patients On Dialysis: Thirteen clinical studies were conducted, involving intravenous administration to a total of 1,010 anemic patients on dialysis for 986 patient-years of PROCRIT therapy. In the three largest of these clinical trials, the median maintenance dose necessary to maintain the hematocrit between 30-36% was approximately 75 Units/kg (T.I.W.). In the U.S. multicenter Phase III study, approximately 65% of the patients required doses of 100 Units/kg (T.I.W.), or less, to maintain their hematocrit at approximately 35%. Almost 10% of patients required a dose of 25 Units/kg, or less, and approximately 10% required a dose of more than 200 Units/kg (T.I.W.) to maintain their hematocrit at this level.

A multicenter unit dose study was also conducted in 119 patients receiving peritoneal dialysis who self-administered PROCRIT subcutaneously for approximately 109 patient-years of experience. Patients responded to PROCRIT administered subcutaneously in a manner similar to patients receiving intravenous administration.[15]

Patients With CRF Not Requiring Dialysis: Four clinical trials were conducted in patients with CRF not on dialysis involving 181 patients treated with PROCRIT for approximately 67 patient-years of experience. These patients responded to PROCRIT therapy in a manner similar to that observed in patients on dialysis. Patients with CRF not on dialysis demonstrated a dose-dependent and sustained increase in hematocrit when PROCRIT was administered by either an intravenous (IV) or subcutaneous (SC) route, with similar rates of rise of hematocrit when PROCRIT was administered by either route. Moreover, PROCRIT doses of 75–150 Units/kg per week have been shown to maintain hematocrits of 36-38% for up to six months. Correcting the anemia of progressive renal failure will allow patients to remain active even though their renal function continues to decrease.[16–18]

Zidovudine-treated HIV-Infected Patients

PROCRIT has been studied in four placebo-controlled trials enrolling 297 anemic (hematocrit < 30%) HIV-infected (AIDS) patients receiving concomitant therapy with zidovudine, (all patients were treated with Epoetin alfa manufactured by Amgen Inc.). In the subgroup of patients (89/125 PROCRIT, and 88/130 placebo) with prestudy endogenous serum erythropoietin levels ≤ 500 mUnits/mL (normal endogenous serum erythropoietin levels are 4–26 mUnits/mL), PROCRIT reduced the mean cumulative number of units of blood transfused per patient by approximately 40%, as compared to the placebo group.[19] Among those patients who required transfusions at baseline, 43% of patients treated with PROCRIT versus 18% of placebo-treated patients were transfusion-independent during the second and third months of therapy. PROCRIT therapy also resulted in significant increases in hematocrit in comparison to placebo. When examining the results according to the weekly dose of zidovudine received during Month 3 of therapy, there was a statistically significant ($p < 0.003$) reduction in transfusion requirements in patients treated with PROCRIT (N=51) compared to placebo-treated patients (N=54) whose mean weekly zidovudine dose was ≤ 4,200 mg/week.[19] Approximately 17% of the patients with endogenous serum erythropoietin levels ≤ 500 mUnits/mL receiving PROCRIT in doses from 100–200 Units/kg three times weekly (T.I.W.) achieved a hematocrit of 38% without administration of transfusions or a significant reduction in zidovudine dose. In the subgroup of patients whose prestudy endogenous serum erythropoietin levels were > 500 mUnits/mL, PROCRIT therapy did not reduce transfusion requirements or increase hematocrit, compared to the corresponding responses in placebo-treated patients.

In a six month open label PROCRIT study, patients responded with decreased transfusion requirements and sustained increases in hematocrit and hemoglobin with doses of PROCRIT up to 300 Units/kg (T.I.W.).[18–20]

Responsiveness to PROCRIT therapy may be blunted by intercurrent infectious/inflammatory episodes and by an increase in zidovudine dosage. Consequently, the dose of PROCRIT must be titrated based on these factors to maintain the desired erythropoietic response.

Cancer Patients on Chemotherapy

PROCRIT has been studied in a series of placebo-controlled, double-blind trials in a total of 131 anemic cancer patients. Within this group, 72 patients were treated with concomitant noncisplatin-containing chemotherapy regimens and 59 patients were treated with concomitant cisplatin-containing chemotherapy regimens. Patients were randomized to PROCRIT 150 Units/kg or placebo subcutaneously (T.I.W.) for 12 weeks.

PROCRIT therapy was associated with a significantly ($p < 0.008$) greater hematocrit response than in the corresponding placebo-treated patients (see TABLE).[19]

HEMATOCRIT (%): MEAN CHANGE FROM BASELINE TO FINAL VALUE[a]

STUDY	PROCRIT	PLACEBO
Chemotherapy	7.6	1.3
Cisplatin	6.9	0.6

[a] Significantly higher in PROCRIT patients than in placebo patients ($p < 0.008$)

In the two types of chemotherapy studies [utilizing a PROCRIT dose of 150 Units/kg (T.I.W.)]the mean number of units of blood transfused per patient after the first month of therapy was significantly ($p < 0.02$) lower in patients treated with PROCRIT (0.71 units in Months 2, 3) than in corresponding placebo-treated patients (1.84 units in Months 2, 3). Moreover, the proportion of patients transfused during Months 2 and 3 of therapy combined was significantly ($p < 0.03$) lower in the patients treated with PROCRIT than in the corresponding placebo-treated patients (22% versus 43%).[19]

Comparable intensity of chemotherapy in the PROCRIT and placebo groups in the chemotherapy trials was suggested by a similar area under the neutrophil time curve in patients treated with PROCRIT and placebo-treated patients as well as by a similar proportion of patients in groups treated with PROCRIT and placebo-treated groups whose absolute neutrophil counts fell below 1,000 cells/μL. Available evidence suggests that patients with lymphoid and solid cancers respond equivalently to PROCRIT therapy, and that patients with or without tumor infiltration of the bone marrow respond equivalently to PROCRIT therapy.

CONTRAINDICATIONS

PROCRIT is contraindicated in patients with:

1) Uncontrolled hypertension.
2) Known hypersensitivity to mammalian cell-derived products.
3) Known hypersensitivity to Albumin (Human).

WARNINGS

Chronic Renal Failure Patients

Hypertension: Patients with uncontrolled hypertension should not be treated with PROCRIT; blood pressure should be controlled adequately before initiation of therapy. Up to 80% of patients with CRF have a history of hypertension.[21] Although there does not appear to be any direct pressor effects of PROCRIT, blood pressure may rise during PROCRIT therapy. During the early phase of treatment when the hematocrit is increasing, approximately 25% of patients on dialysis may require initiation of, or increases in, antihypertensive therapy. Hypertensive encephalopathy and seizures have been observed in patients with CRF treated with PROCRIT.

Special care should be taken to closely monitor and aggressively control blood pressure in patients treated with PROCRIT. Patients should be advised as to the importance of compliance with antihypertensive therapy and dietary restrictions. If blood pressure is difficult to control by initiation of appropriate measures, the hematocrit may be reduced by decreasing or withholding the dose of PROCRIT. A clinically significant decrease in hematocrit may not be observed for several weeks.

It is recommended that the dose of PROCRIT be decreased if the hematocrit increase exceeds 4 points in any two-week period, because of the possible association of excessive rate of rise of hematocrit with an exacerbation of hypertension.

Seizures: Seizures have occurred in patients with CRF participating in PROCRIT clinical trials.

In patients on dialysis, there was a higher incidence of seizures during the first 90 days of therapy (occurring in approximately 2.5% of patients) as compared with later time-points.

Given the potential for an increased risk of seizures during the first 90 days of therapy, blood pressure and the presence of premonitory neurologic symptoms should be monitored closely. Patients should be cautioned to avoid potentially hazardous activities such as driving or operating heavy machinery during this period.

While the relationship between seizures and the rate of rise of hematocrit is uncertain, it is recommended that the dose of PROCRIT be decreased if the hematocrit increase
4 points in any two-week period.

Thrombotic Events: During hemodialysis, patients with PROCRIT may require increased anticoagula heparin to prevent clotting of the artificial kidney tionship has not been established with statistical between a rise in hematocrit and the rate of th events (including thrombosis of vascular access). In trials, clotting of the vascular access (A-V shunt curred at an annualized rate of about 0.25 events pe year on PROCRIT therapy, a rate which appears higher than that seen in untreated patients on Overall, for patients with CRF (whether on dialys other thrombotic events (e.g., myocardial infarction vascular accident, transient ischemic attack) have in clinical trials at an annualized rate of less events per patient-year of PROCRIT therapy. Pa pre-existing vascular disease should be monitor See "ADVERSE REACTIONS" for more inform thrombotic events.

Zidovudine-treated HIV-Infected Patients

In contrast to CRF patients, PROCRIT therapy ha linked to exacerbation of hypertension, seizures, botic events in HIV-infected patients.

Miscellaneous: The multidose preserved formu tains benzyl alcohol. Benzyl alcohol has been repo associated with an increased incidence of neurolo other complications in premature infants which times fatal.

PRECAUTIONS

Chronic Renal Failure Patients, Zidovudine-trea fected Patients and Cancer Patients on Chemoth

General: The parenteral administration of any bio uct should be attended by appropriate precautio allergic or other untoward reactions occur (see cations"). In clinical trials, while transient rashes sionally observed concurrently with PROCRIT th serious allergic or anaphylactic reactions were rep "ADVERSE REACTIONS" for more information allergic reactions.

The safety and efficacy of PROCRIT therapy ha established in patients with a known history disorder or underlying hematologic disease (e anemia, myelodysplastic syndromes, or hyp disorders).

In some female patients, menses have resumed PROCRIT therapy; the possibility of pregnancy discussed and the need for contraception evalu

Hematology: Exacerbation of porphyria observed rarely in patients with CRF PROCRIT. However, PROCRIT has not caused urinary excretion of porphyrin metabolites in no teers, even in the presence of a rapid eryth sponse. Nevertheless, PROCRIT should be used in patients with known porphyria.

In pre clinical studies in dogs and rats, but no PROCRIT therapy was associated with subclini row fibrosis. Bone marrow fibrosis is a known of CRF in humans and may be related to secon parathyroidism or unknown factors. The incide marrow fibrosis was not increased in a study of dialysis who were treated with PROCRIT for 1 compared to the incidence of bone marrow matched group of patients who had not been PROCRIT.

Hematocrit in CRF patients should be meas week; zidovudine-treated HIV-infected and ca should have hematocrit measured once a week crit has been stabilized, and measured periodi ter.

Delayed or Diminished Response: If the respond or to maintain a response to doses wi mended dosing range, the following etiologies sidered and evaluated:

1) Iron deficiency: Virtually all patients will quire supplemental iron therapy. (See "Iron
2) Underlying infectious, inflammatory, or cesses.
3) Occult blood loss.
4) Underlying hematologic diseases (i.e., thala tory anemia, or other myelodysplastic dis
5) Vitamin deficiencies: folic acid or vitamin
6) Hemolysis.
7) Aluminum intoxication.
8) Osteitis fibrosa cystica.

Iron Evaluation: During PROCRIT therapy functional iron deficiency may develop. Func ciency, with normal ferritin levels but low ration, is presumably due to the inability stores rapidly enough to support increased Transferrin saturation should be at least 2 should be at least 100 ng/mL.

Prior to and during PROCRIT therapy, the status, including transferrin saturation (ser by iron binding capacity) and serum ferritin

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Event	PERCENT OF PATIENTS REPORTING EVENT: Patients Treated with PROCRIT (N=200)	PLACEBO-TREATED Patients (N=135)
Hypertension	24%	19%
Headache	16%	12%
Arthralgias	11%	6%
Nausea	11%	9%
Edema	9%	10%
Fatigue	9%	14%
Diarrhea	9%	6%
Vomiting	8%	5%
Chest Pain	7%	9%
Skin Reaction (Administration Site)	7%	12%
Asthenia	7%	12%
Dizziness	7%	13%
Clotted Access	7%	2%
	1.1%	1.1%
	0.4%	0.6%
	0.4%	1.1%
	0	1.7%

rus, and potassium]should be monitored regularly. During clinical trials in patients on dialysis, modest increases were seen in BUN, creatinine, phosphorus, and potassium. In some patients with CRF not on dialysis, treated with PROCRIT, modest increases in serum uric acid and phosphorus were observed. While changes were statistically significant, the values remained within the ranges normally seen in patients with CRF.

Diet: As the hematocrit increases and patients experience an improved sense of well-being and quality of life, the importance of compliance with dietary and dialysis prescriptions should be reinforced. In particular, hyperkalemia is not uncommon in patients with CRF. In U.S. studies in patients on dialysis, hyperkalemia has occurred at an annualized rate of approximately 0.11 episodes per patient-year of PROCRIT therapy, often in association with poor compliance to medication, dietary and/or dialysis prescriptions.

Dialysis Management: Therapy with PROCRIT results in an increase in hematocrit and a decrease in plasma volume which could affect dialysis efficiency. In studies to date, the resulting increase in hematocrit did not appear to adversely affect dialyzer function[9,10] or the efficiency of high flux hemodialysis.[11] During hemodialysis, patients treated with PROCRIT may require increased anticoagulation with heparin to prevent clotting of the artificial kidney.

Patients who are marginally dialyzed may require adjustments in their dialysis prescription. As with all patients on dialysis, the serum chemistry values [including blood urea nitrogen (BUN), creatinine, phosphorus, and potassium] in patients treated with PROCRIT should be monitored regularly to assure the adequacy of the dialysis prescription.

Information for Patients: In those situations in which the physician determines that a home dialysis patient can safely and effectively self-administer PROCRIT, the patient should be instructed as to the proper dosage and administration. Home dialysis patients should be referred to the full "Information for Home Dialysis Patients" section attached; it is not a disclosure of all possible effects. Patients should be informed of the signs and symptoms of allergic drug reaction and advised of appropriate actions. If home use is prescribed for a home dialysis patient, the patient should be thoroughly instructed in the importance of proper disposal and cautioned against the reuse of needles, syringes, or drug product. A puncture-resistant container for the disposal of used syringes and needles should be available to the patient. The full container should be disposed of according to the directions provided by the physician.

Renal Function: In patients with CRF not on dialysis, renal function and fluid and electrolyte balance should be closely monitored, as an improved sense of well-being may obscure the need to initiate dialysis in some patients. In patients with CRF not on dialysis, placebo-controlled studies of progression of renal dysfunction over periods of greater than one year have not been completed. In shorter-term trials in patients with CRF not on dialysis, changes in creatinine and creatinine clearance were not significantly different in patients treated with PROCRIT, compared with placebo-treated patients. Analysis of the slope of 1/serum creatinine vs. time plots in these patients indicates no significant change in the slope after the initiation of PROCRIT therapy.

Zidovudine-treated HIV-Infected Patients

Hypertension: Exacerbation of hypertension has not been observed in zidovudine-treated HIV-infected patients treated with PROCRIT. However, PROCRIT should be withheld in these patients if pre-existing hypertension is uncontrolled, and should not be started until blood pressure is controlled. In double-blind studies, a single seizure has been experienced by a patient treated with PROCRIT.[19]

Cancer Patients on Chemotherapy

Hypertension: Hypertension, associated with a significant increase in hematocrit, has been noted rarely in cancer patients treated with PROCRIT. Nevertheless, blood pressure in patients treated with PROCRIT should be monitored carefully, particularly in patients with an underlying history of hypertension or cardiovascular disease.

Seizures: In double-blind, placebo-controlled trials, 3.2% (N=2/63) of patients treated with PROCRIT and 2.9% (N=2/68) of placebo-treated patients had seizures. Seizures in 1.6% (N=1/63) of patients treated with PROCRIT occurred in the context of a significant increase in blood pressure and hematocrit from baseline values. However, both patients treated with PROCRIT also had underlying CNS pathology which may have been related to seizure activity.

Thrombotic Events: In double-blind, placebo-controlled trials, 3.2% (N=2/63) of patients treated with PROCRIT and 11.8% (N=8/68) of placebo-treated patients had thrombotic events (e.g. pulmonary embolism, cerebrovascular accident).

Growth Factor Potential: PROCRIT is a growth factor that primarily stimulates red cell production. However, the possibility that PROCRIT can act as a growth factor for any tumor type, particularly myeloid malignancies, cannot be excluded.

ADVERSE REACTIONS

Chronic Renal Failure Patients

Studies analyzed to date indicate that PROCRIT is generally well-tolerated. The adverse events reported are frequent sequelae of CRF and are not necessarily attributable to PROCRIT therapy. In double-blind, placebo-controlled studies involving over 300 patients with CRF, the events reported in greater than 5% of patients treated with PROCRIT during the blinded phase were:

[See table above.]

Significant adverse events of concern in patients with CRF treated in double-blinded, placebo-controlled trials occurred in the following percent of patients during the blinded phase of the studies:

[See table below.]

In the U.S. PROCRIT studies in patients on dialysis (over 567 patients), the incidence (number of events per patient-year) of the most frequently reported adverse events were: hypertension (0.75), headache (0.40), tachycardia (0.31), nausea/vomiting (0.26), clotted vascular access (0.25), shortness of breath (0.14), hyperkalemia (0.11), and diarrhea (0.11). Other reported events occurred at a rate of less than 0.10 events per patient per year.

Events reported to have occurred within several hours of administration of PROCRIT were rare, mild, and transient, and included injection site stinging in dialysis patients and flu-like symptoms such as arthralgias and myalgias.

In all studies analyzed to date, PROCRIT administration was generally well-tolerated, irrespective of the route of administration.

Hypertension: Increases in blood pressure have been reported in clinical trials, often during the first 90 days of therapy. On occasion, hypertensive encephalopathy and seizures have been observed in patients with CRF treated with PROCRIT. When data from all patients in the U.S. Phase III multicenter trial were analyzed, there was an apparent trend of more reports of hypertensive adverse events in patients on dialysis with a faster rate of rise of hematocrit (greater than 4 hematocrit points in any two-week period). However, in a double-blind, placebo-controlled trial, hypertensive adverse events were not reported at an increased rate in the group treated with PROCRIT (150 Units/kg (T.I.W.) relative to the placebo group.

Continued on next page

Ortho Biotech—Cont.

Seizures: There have been 47 seizures in 1,010 patients on dialysis treated with PROCRIT in clinical trials, with an exposure of 986 patient-years for a rate of approximately 0.048 events per patient-year. However, there appeared to be a higher rate of seizures during the first 90 days of therapy (occurring in approximately 2.5% of patients) when compared to subsequent 90-day periods. The baseline incidence of seizures in the untreated dialysis population is difficult to determine; it appears to be in the range of 5-10% per patient-year.[22-24]

Thrombotic Events: In clinical trials, clotting of the vascular access has occurred at an annualized rate of about 0.25 events per patient-year on PROCRIT therapy. Overall, for patients with CRF (whether on dialysis or not), other thrombotic events (e.g., myocardial infarction, cerebrovascular accident, transient ischemic attack) have occurred at an annualized rate of less than 0.04 events per patient-year of PROCRIT therapy.

In over 125,000 patients treated with commercial PROCRIT, there have been rare reports of serious or unusual thrombo-embolic events including migratory thrombophlebitis, microvascular thrombosis pulmonary embolus, and thrombosis of the retinal artery, and temporal and renal veins. Collectively, these events have been reported in < 0.0001 events per patient-year; in no case has a casual relationship been established.

Allergic Reactions: There have been no reports of serious allergic reactions or anaphylaxis associated with PROCRIT administration during clinical trials. Skin rashes and urticaria have been observed rarely and when reported have generally been mild and transient in nature.

In over 125,000 patients treated with commercial PROCRIT, there have been rare reports of potentially serious allergic reactions including urticaria with associated respiratory symptoms or circumoral edema (< 0.0001 events per patient-year), or urticaria alone (< 0.0001 events per patient-year). Most reactions occurred in situations where a casual relationship could not be established. Many of these patients resumed PROCRIT therapy without recurrence of symptoms, some in conjunction with antihistamine pretreatment. However, symptoms recurred with rechallenge in a few instances, suggesting that allergic reactivity, although rare, may occasionally be associated with PROCRIT therapy. There has been no evidence for development of antibodies to erythropoietin in patients tested to date, including those receiving PROCRIT for over 4 years. Nevertheless, if an anaphylactoid reaction occurs, PROCRIT should be immediately discontinued and appropriate therapy initiated.

Zidovudine-treated HIV-infected Patients

Adverse events reported in clinical trials with PROCRIT in zidovudine-treated HIV-infected patients were consistent with the progression of HIV infection. In double-blind, placebo-controlled studies of three-months duration involving approximately 300 zidovudine-treated HIV-infected patients, adverse events with an incidence of ≥ 10% in either patients treated with PROCRIT or placebo-treated patients were:

[See table below.]

There were no statistically significant differences between treatment groups in the incidence of the above events.

In the 297 patients studied, PROCRIT was not associated with significant increases in opportunistic infections or mortality.[19] In 71 patients from this group treated with PROCRIT at 150 Units/kg (T.I.W.), serum p24 antigen levels did not appear to increase.[20] Preliminary data showed no enhancement of HIV replication in infected cell lines *in vitro*.[19]

Peripheral white blood cell and platelet counts are unchanged following PROCRIT therapy.

Allergic Reactions: Two zidovudine-treated HIV-infected patients had urticarial reactions within 48 hours of their first exposure to study medication. One patient was treated with PROCRIT and one was treated with placebo (PROCRIT vehicle alone). Both patients had positive immediate skin tests against their study medication with a negative saline control. The basis for this apparent pre-existing hypersensitivity to components of the PROCRIT formulation is unknown, but may be related to HIV-induced immunosuppression or prior exposure to blood products.

Seizures: In double-blind and open label trials of PROCRIT in zidovudine-treated HIV-infected patients, ten patients have experienced seizures.[19] In general, these seizures appear to be related to underlying pathology such as meningitis or cerebral neoplasms, not PROCRIT therapy.

Cancer Patients on Chemotherapy

Adverse experiences reported in clinical trials with PROCRIT in cancer patients were consistent with the underlying disease state. In double-blind, placebo-controlled studies of up to 3-months duration involving 131 cancer patients, adverse events with an incidence > 10% in either patients treated with PROCRIT or placebo-treated patients were as indicated below.

[See table above.]

Although some statistically significant differences between patients treated with PROCRIT and placebo-treated patients were noted, the overall safety profile of PROCRIT appeared to be consistent with the disease process of advanced cancer.

During double-blind and subsequent open-label therapy in which patients (N = 72 for total exposure to PROCRIT) were treated for up to 32 weeks with doses as high as 927 Units/kg, the adverse experience profile of PROCRIT was consistent with the progression of advanced cancer.

Based on comparable survival data and on the percentage of patients treated with PROCRIT and placebo-treated patients who discontinued therapy due to death, disease progression or adverse experiences (22% and 13%, respectively; p = 0.25), the clinical outcome in patients treated with PROCRIT and placebo-treated patients appeared to be similar. Available data from animal tumor models and measurement of proliferation of solid tumor cells from clinical biopsy specimens in response to PROCRIT suggest that PROCRIT does not potentiate tumor growth. Nevertheless, as a growth factor, the possibility that PROCRIT may potentiate growth of some tumors, particularly myeloid tumors, cannot be excluded. A randomized controlled Phase IV study is currently ongoing to further evaluate this issue.

The mean peripheral white blood cell count was unchanged following PROCRIT therapy compared to the corresponding value in the placebo-treated group.

Percent of Patients Reporting Event

Events	Patients Treated with PROCRIT (N = 63)	PLACEBO-Treated Patients (N = 68)
Pyrexia	29%	19%
Diarrhea	21%[a]	7%
Nausea	17%[b]	32%
Vomiting	17%	15%
Edema	17%[c]	1%
Astheria	13%	16%
Fatigue	13%	15%
Shortness of Breath	13%	9%
Paresthesia	11%	6%
Upper Respiratory Infection	11%	4%
Dizziness	5%	12%
Trunk Pain	3%[d]	16%

[a] p = 0.041
[b] p = 0.069
[c] p = 0.0016
[d] p = 0.017

Percent of Patients Reporting Event

Event	Patients Treated with PROCRIT (N = 144)	PLACEBO-Treated Patients (N = 153)
Pyrexia	38%	29%
Fatigue	25%	31%
Headache	19%	14%
Cough	18%	14%
Diarrhea	16%	18%
Rash	16%	8%
Congestion, Respiratory	15%	10%
Nausea	15%	12%
Shortness of Breath	14%	13%
Asthenia	11%	14%
Skin Reaction, (Administration Site)	10%	7%
Dizziness	9%	10%

OVERDOSAGE

The maximum amount of PROCRIT that can be administered in single or multiple doses has not been determined. Doses of up to 1,500 Units/kg (T.I.W.) for three weeks have been administered without any direct effects of PROCRIT itself.[6] Therapy with PROCRIT in polycythemia if the hematocrit is not carefully and the dose appropriately adjusted. If the suggested range is exceeded, PROCRIT may be temporarily until the hematocrit returns to the suggested range. PROCRIT therapy may then be resumed using a (see "Dosage and Administration"). If polycythemia concern, phlebotomy may be indicated to decrease matocrit.

DOSAGE AND ADMINISTRATION

Chronic Renal Failure Patients

Starting doses of PROCRIT over the range of 50 kg three times weekly (T.I.W.) have been shown and effective in increasing hematocrit and transfusion dependency in patients with CRF (Experience"). The dose of PROCRIT should be hematocrit approaches 36% or increases by more points in any 2-week period. The dosage of PROCRIT individualized to maintain the hematocrit within gested target range. At the physician's discretion, gested target hematocrit range may be expanded to maximal patient benefit.

PROCRIT may be given either as an intravenous (IV) cutaneous (SC) injection. In patients on hemo PROCRIT usually has been administered via a (T.I.W.). While the administration of PROCRIT is dent of the dialysis procedure, PROCRIT may be tered into the venous line at the end of the dialysis to obviate the need for additional venous access. with CRF not on dialysis, PROCRIT may be given an IV or SC injection.

Home hemodialysis patients who have been judged tent by their physicians to self-administer PROCRIT medical or other supervision may give themselves IV or SC injection. Home peritoneal dialysis have been judged competent by their physicians minister PROCRIT without medical or other may give themselves a SC injection. The table vides general therapeutic guidelines for patient

[See table at top of next page.]

During therapy, hematological parameters tored regularly (see "Laboratory Monitoring").

Pre-Therapy Iron Evaluation: Prior to PROCRIT therapy, the patient's iron stores, ferrin saturation (serum iron divided by ity) and serum ferritin, should be evaluated saturation should be at least 20%, and ferritin least 100 ng/mL. Virtually all patients will quire supplemental iron to increase or maintain saturation to levels that will adequately support esis stimulated by PROCRIT.

Dose Adjustment: Following PROCRIT therapy time is required for erythroid progenitors to mature released into circulation resulting in an increase hematocrit. Additionally, red blood cell survival hematocrit and may vary due to uremia. As a result required to elicit a clinically significant change crit (increase or decrease) following any dose may be 2–6 weeks.

Dose adjustment should not be made more frequently once a month, unless clinically indicated. After adjustment, the hematocrit should be determined weekly for at least 2–6 weeks (see "Laboratory Mon

...atocrit is increasing and approaching 36%, the ...ld be reduced to maintain the suggested target he... range. If the reduced dose does not stop the rise in ... and it exceeds 36%, doses should be temporarily ... until the hematocrit begins to decrease, at which ...apy should be reinitiated at a lower dose.
...time, if the hematocrit increases by more than 4 ... 2-week period, the dose should be immediately ... After the dose reduction, the hematocrit should ...ed twice weekly for 2–6 weeks, and further dose ...ts should be made as outlined in "Maintenance

...atocrit increase of 5–6 points is not achieved after ... period and iron stores are adequate ...ayed or Diminished Response"), the dose of ... may be incrementally increased. Further in... may be made at 4–6 week intervals until the desired ... is attained.

...nce Dose: The maintenance dose must be individ... for each patient on dialysis. In the U.S. Phase III ... trial in patients on hemodialysis, the median ...nce dose was 75 Units/kg (T.I.W.), with a range ... to 525 Units/kg (T.I.W.). Almost 10% of the pa...quired a dose of 25 Units/kg, or less, and approxi...% of the patients required more than 200 Units/kg ... maintain their hematocrit in the suggested target

...atocrit remains below, or falls below, the sug...get range, iron stores should be re-evaluated. If the ... saturation is less than 20%, supplemental iron ... administered. If the transferrin saturation is ...an 20%, the dose of PROCRIT may be increased. ... increases should not be made more frequently ... a month, unless clinically indicated, as the re...se of the hematocrit to a dose increase can be 2–6 ...ematocrit should be measured twice weekly for 2–6 ...owing dose increases. In patients with CRF not on ... the maintenance dose must also be individualized. ...ll doses of 75–150 Units/kg <u>per week</u> have been ... maintain hematocrits of 36–38% for up to 6

...Diminished Response: Over 95% of patients ...sponded with clinically significant increases in ... virtually all patients were transfusion-inde... approximately two months of initiation of ...therapy.

... fails to respond or maintain a response, other ... should be considered and evaluated as clinically ... the "PRECAUTIONS" section for discussion of ... diminished response.

...treated HIV-Infected Patients

... beginning PROCRIT, it is recommended that the ... serum erythropoietin level be determined (prior ...). Available evidence suggests that patients ... zidovudine with endogenous serum erythropoietin ... mUnits/mL are unlikely to respond to therapy ...CRIT.

...Dose: For patients with serum erythropoietin ... mUnits/mL who are receiving a dose of zidovu... mg/week, the recommended starting dose of ... 100 Units/kg as an intravenous or subcutane... three times weekly (T.I.W.) for 8 weeks.

...Dose: During the dose adjustment phase of ther... hematocrit should be monitored weekly. If the re... not satisfactory in terms of reducing transfusion ... or increasing hematocrit after 8 weeks of ther... dose of PROCRIT can be increased by 50–100 Units/... Response should be evaluated every 4–8 weeks ... and the dose adjusted accordingly by 50–100 ... increments (T.I.W.). If patients have not responded ... to a PROCRIT dose of 300 Units/kg (T.I.W.), it ... that they will respond to higher doses of

...Dose: After attainment of the desired re... reduced transfusion requirements or increased ... the dose of PROCRIT should be titrated to main... response based on factors such as variations in ... dose and the presence of intercurrent infectious ...atory episodes. If the hematocrit exceeds 40%, ... should be discontinued until the hematocrit drops to ... dose should be reduced by 25% when treatment is ... and then titrated to maintain the desired hemato...

...on Chemotherapy

...ndogenous serum erythropoietin levels varied ... in these trials with approximately 75% ... having endogenous serum erythropoietin levels ...mU/mL, and approximately 4% (N=4/110) of ... having endogenous serum erythropoietin levels ...mU/mL. In general, patients with lower baseline ...opoietin levels responded more vigorously to ... than patients with higher erythropoietin levels. ... specific serum erythropoietin level can be stipu... which patients would be unlikely to respond to ... therapy, treatment of patients with grossly ele... erythropoietin levels (e.g., > 200 mUnits/mL) is

Starting Dose	Reduce Dose When	Increase Dose If	Maintenance Dose	Suggested Target Hct. Range
50–100 Units/kg T.I.W.; IV or SC	1) Hct. approaches 36%, or 2) Hct. increases > 4 points in any 2-week period	Hct. does not increase by 5–6 points after 8 weeks of therapy, and hct. is below suggested target range	Individually titrate	30–36%

BEST AVAILABLE COPY

not recommended. The hematocrit should be monitored on a weekly basis in patients receiving PROCRIT therapy until hematocrit becomes stable.

Starting Dose: The recommended starting dose of PROCRIT is 150 Units/kg subcutaneously (T.I.W.).

Dose Adjustment: If the response is not satisfactory in terms of reducing transfusion requirements or increasing hematocrit after 8 weeks of therapy, the dose of PROCRIT can be increased up to 300 Units/kg (T.I.W.). If patients have not responded satisfactorily to a PROCRIT dose of 300 Units/kg (T.I.W.), it is unlikely that they will respond to higher doses of PROCRIT. If the hematocrit exceeds 40%, the dose of PROCRIT should be withheld until the hematocrit falls to 36%. The dose of PROCRIT should be reduced by 25% when treatment is resumed and titrated to maintain the desired hematocrit. If the initial dose of PROCRIT includes a very rapid hematocrit response (e.g., an increase of more than 4 percentage points in any 2-week period), the dose of PROCRIT should be reduced.

PREPARATION AND ADMINISTRATION OF PROCRIT

1. DO NOT SHAKE. It is not necessary to shake PROCRIT. Prolonged vigorous shaking may denature any glycoprotein, rendering it biologically inactive.
2. Parenteral drug products should be inspected visually for particulate matter and discoloration prior to administration. Do not use any vials exhibiting particulate matter or discoloration.
3. Using aseptic techniques, attach a sterile needle to a sterile syringe. Remove the flip top from the vial containing PROCRIT, and wipe the septum with a disinfectant. Insert the needle into the vial, and withdraw into the syringe an appropriate volume of solution.
4. **Single-dose** 1 mL vial contains no preservative. Use one dose per vial; do not re-enter vial. Discard unused portions. **Multidose** 2 mL vial contains preservative. Store at 2 to 8°C after initial entry and between doses. Discard 21 days after initial entry.
5. Do not dilute or administer in conjunction with other drug solutions. However, at the time of subcutaneous administration, PROCRIT may be admixed in a syringe with bacteriostatic 0.9% sodium chloride injection, USP, with benzyl alcohol 0.9% (bacteriostatic saline) at a 1:1 ratio using aseptic technique. The benzyl alcohol in the bacteriostatic saline acts as a local anesthetic which may ameliorate subcutaneous injection site discomfort.

HOW SUPPLIED

PROCRIT, containing Epoetin alfa, is available in vials containing color coded labels.

1 mL Single-Dose, Preservative-Free Solution

Each dosage form is supplied in the following packages:

Cartons containing six (6) **single-dose** vials:
2,000 Units/mL (NDC 59676-302-01) (Purple)
3,000 Units/mL (NDC 59676-303-01) (Magenta)
4,000 Units/mL (NDC 59676-304-01) (Green)
10,000 Units/mL (NDC 59676-310-01) (Red)

Trays containing twenty-five (25) **single-dose** vials:
2,000 Units/mL (NDC 59676-302-02) (Purple)
3,000 Units/mL (NDC 59676-303-02) (Magenta)
4,000 Units/mL (NDC 59676-304-02) (Green)
10,000 Units/mL (NDC 59676-310-02) (Red)

2 mL Multidose, Preserved Solution

Cartons containing six (6) multidose vials:
10,000 Units/mL (NDC 59676-312-01) (Blue)

STORAGE

Store at 2° to 8° C (36° to 46° F). Do not freeze or shake.

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Distributed by:
Ortho Biotech Inc.
Raritan, New Jersey 08869-0670

Revised May 1995
Printed in U.S.A.

PROCRIT®
EPOETIN ALFA ℞

INFORMATION FOR HOME DIALYSIS PATIENTS
PROCRIT and Chronic Renal Failure

PROCRIT (Epoetin alfa) has been prescribed for you by your doctor because you:

1) Have anemia due to your kidney disease.
2) Are able to dialyze at home.
3) Have been determined to be able to administer PROCRIT without direct medical or other supervision.

A lack of energy or feeling of tiredness is the major symptom of anemia. Additional symptoms include shortness of breath, chest pain, and feeling cold all the time. The reason for these symptoms is that there is a lack of red blood cells. Red blood cells carry oxygen, which is important for all of the body's functions. When there are fewer red blood cells, the body does not get all the oxygen it needs.

Kidneys remove toxins from the blood; they also measure the amount of oxygen in the blood. If there is not enough oxygen, the kidneys will produce a hormone called erythropoietin. Erythropoietin is released into the bloodstream and travels to the bone marrow where red blood cells are made. Erythropoietin signals the bone marrow to make more oxygen-carrying red blood cells.

As the kidneys fail, they stop cleansing toxins from your body. They also make less erythropoietin than they should. Therefore, the bone marrow does not receive a strong-enough signal to make the oxygen-carrying red blood cells. Fewer red blood cells are produced so the muscles, brain, and other parts of the body do not get the oxygen they need to function properly.

PROCRIT is a copy of human erythropoietin. PROCRIT replaces the erythropoietin that the failed kidneys can no longer produce, and signals the bone marrow to make the oxygen-carrying red blood cells once again.

The effectiveness of PROCRIT is measured by the increase in hematocrit (the amount of red blood cells in the blood) that results from PROCRIT therapy. The rise in hematocrit is not immediate. It usually takes about two to six weeks before the hematocrit starts to rise. The amount of time it takes, and the dose of PROCRIT that is needed to make the hematocrit increase, varies from patient to patient.

Most patients treated with PROCRIT no longer need blood transfusions. However, certain medical conditions, or unexpected blood loss, may result in the need for a transfusion.

In those situations where your doctor has determined that you, as a home dialysis patient, can self-administer PROCRIT, you will receive instruction on how much PROCRIT to use, how to inject it, how often you should inject it, and how you should dispose of the unused portions of each vial.

You will be instructed to monitor your blood pressure carefully every day and to report any changes outside of the guidelines that your doctor has given you. When the number of red blood cells increases, your blood pressure can also increase, so your doctor may prescribe some new or additional blood pressure medication. Be sure to follow your doctor's orders. You may also be instructed to have certain laboratory tests, such as additional hematocrit or iron level measurements, done more frequently. You may be asked to report these tests to your doctor or dialysis center. Also, your doctor may prescribe additional iron for you to take. Be sure to comply with your doctor's orders.

Continue to check your access, as your doctor or nurse has shown you, to make sure it is working. Be sure to let your health care professional know right away if there is a problem.

When you receive your PROCRIT from the dialysis center, doctor's office or home dialysis supplier, always check to see that:

1) The name PROCRIT appears on the carton and bottle label.
2) You will be able to use PROCRIT before the expiration date stamped on the package.

PROCRIT

PROCRIT is produced in mammalian cells that have been genetically altered by the addition of a gene for the natural substance erythropoietin.

The PROCRIT solution in the vial should always be clear and colorless. Do not use PROCRIT if the contents of the vial appear discolored or cloudy, or if the vial appears to contain lumps, flakes, or particles. In addition, if the vial has been shaken vigorously, the solution may appear to be frothy and should not be used. Therefore, care should be taken not to shake the PROCRIT vial vigorously before use.

Single-dose vial: 2000, 3000, 4000 and 10,000 Unit vials of PROCRIT are for single use only. Any unused portions of these vials should be discarded.

Multi-dose Vial: Vials of PROCRIT marked with a blue "M" on the label (10,000 Units/mL- 20,000 Units in a vial) contain a preserved solution and may be entered multiple times. Multidose vials should be stored in the refrigerator between uses and thrown away 21 days after the first use. Carefully follow the instructions for "Preparing the Dose" each time you enter the multidose vial.

Follow your dialysis center's instructions on what to do with the used vials.

Storage:

PROCRIT should be stored in the refrigerator, but not in the freezing compartment. Do not let the vial freeze and do not leave it in direct sunlight. Do not use a vial of PROCRIT that has been frozen or after the expiration date that is stamped on the label. If you have any questions about the safety of a vial of PROCRIT that has been subjected to temperature extremes, be sure to check with your dialysis unit staff.

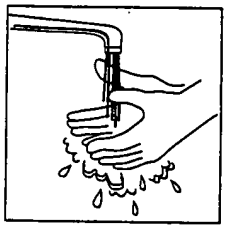
USE THE CORRECT SYRINGE

Your doctor has instructed you on how to give yourself the correct dosage of PROCRIT. This dosage will usually be measured in units per milliliter or ccs. It is important to use a syringe that is marked in tenths of milliliters (for example, 0.1, 0.2, etc., mL or cc). Failure to use the proper syringe can lead to a mistake in dosage, and you may receive too much or too little PROCRIT. Too little PROCRIT may not be effective in increasing your hematocrit, and too much PROCRIT may lead to a hematocrit that is too high. Only use disposable syringes and needles as they do not require sterilization; they should be used once and disposed of as instructed by your doctor.

IMPORTANT: TO HELP AVOID CONTAMINATION AND POSSIBLE INFECTION, FOLLOW THESE INSTRUCTIONS EXACTLY.

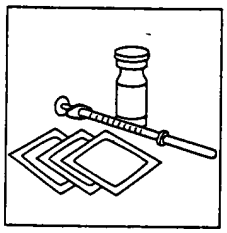
PREPARING THE DOSE:

1. Wash your hands thoroughly with soap and water before preparing the medication.

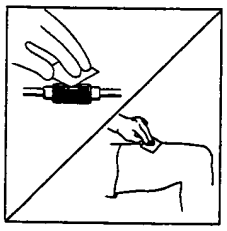


2. Check the date on the PROCRIT vial to be sure that the drug has not expired.

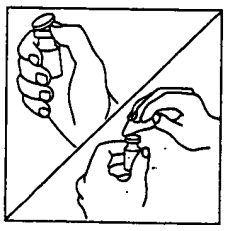
3. Remove the vial of PROCRIT from the refrigerator and allow it to reach room temperature. It is not necessary to shake PROCRIT. Prolonged vigorous shaking may damage the product. Assemble the other supplies you will need for your injection.



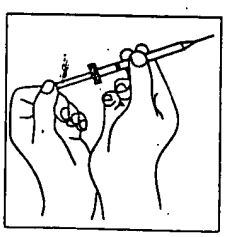
4. Hemodialysis patients should wipe off the venous port of the hemodialysis tubing with an antiseptic swab. Peritoneal dialysis patients should cleanse the skin with an antiseptic swab where the injection is to be made.



5. Flip off the red protective cap but do not remove the gray rubber stopper. Wipe the top of the gray rubber stopper with an antiseptic swab.

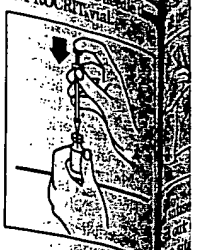


6. Using a syringe and needle designed for subcutaneous injection, draw air into the syringe by pulling back on the plunger. The amount of air should be equal to your PROCRIT dose.

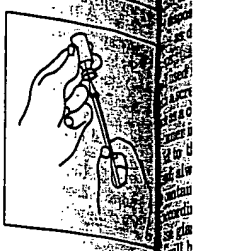


7. Carefully remove the needle cover. Put the needle through the gray rubber stopper of the PROCRIT vial.

8. Push the plunger into discharge air into the vial. The air injected into the vial will allow PROCRIT to be easily withdrawn into the syringe.



9. Turn the vial and syringe upside down in one hand. Be sure the tip of the needle is in the PROCRIT solution. Your other hand will be free to move the plunger. Draw back on the plunger slowly to draw the correct dose of PROCRIT into the syringe.



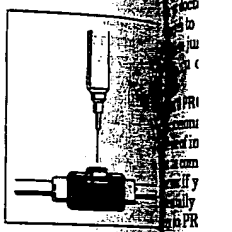
10. Check for air bubbles. The air is harmless, but an air bubble will reduce the PROCRIT dose. To remove bubbles, gently tap the syringe to move the air bubbles to the top of the syringe, then use the plunger to push the air and the air back into the vial. Then re-measure your dose of PROCRIT.

11. Double check your dose. Remove the needle from the vial. Do not lay the syringe down or allow the needle to touch anything.

INJECTING THE DOSE

Patients on home hemodialysis using the intravenous injection route:

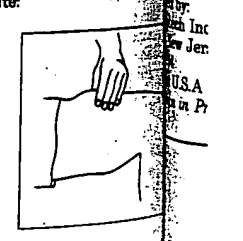
1. Insert the needle of the syringe into the previously cleansed venous port and inject the PROCRIT.



2. Remove the syringe and dispose of the whole unit. **Use disposable syringe only once.** Dispose of syringes and needles as directed by your doctor, by following these simple guidelines:
—Place all used needles and syringes in a hard-plastic container with a screw-on-cap, or a metal container with a plastic lid, such as a coffee can properly labeled as to the hazard. If a metal container is used, cut a small hole in the plastic lid and tape the lid to the metal container. If a hard-plastic container is used, always screw the cap on tightly after each use. When the container is full, tape around the cap or lid and dispose of according to your doctor's instructions.
—Do not use glass or clear plastic containers, or any container that will be recycled or returned to a store.
—Always store the container out of the reach of children.
—Please check with your doctor, nurse, or pharmacist for other suggestions. There may be special state and local guidelines that they will discuss with you.

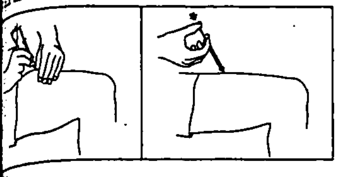
Patients on home peritoneal dialysis or home hemodialysis using the subcutaneous route:

1. With one hand, stabilize the previously cleansed skin by spreading it or by pinching up a large area with your free hand.



2. Hold the syringe with the other hand, as you would a pencil. Double check that the correct amount of PROCRIT is in the syringe. Insert the needle straight into the skin (90 degree angle). Pull the plunger back slightly. If blood comes into the syringe, do not inject PROCRIT; the needle has entered a blood vessel; withdraw the syringe

ifferent site. Inject the PROCRIT by pushing the the way down.



antiseptic swab near the needle and pull the nee- out of the skin. Press the antiseptic swab over site for several seconds.

disposable syringe only once. Dispose of syringes as directed by your doctor, by following these

used needles and syringes in a hard plastic con- a screw-on-cap, or a metal container with a plas- as a coffee can properly labeled as to content. If a iner is used, cut a small hole in the plastic lid and lid to the metal container. If a hard-plastic con- always screw the cap on tightly after each use. container is full, tape around the cap or lid, and according to your doctor's instructions.

use glass or clear plastic containers, or any con- will be recycled or returned to a store.

store the container out of the reach of children.

check with your doctor, nurse, or pharmacist for questions. There may be special state and local laws will discuss with you.

change
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bruising
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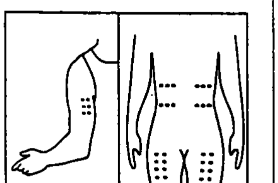
doctor.
to
just
can



TO PROCRIT

occasionally experience redness, swelling, or itch- of injection of PROCRIT. This may indicate an the components of PROCRIT, or it may indicate a If you have a local reaction, consult your doc- ally more serious reaction would be a general- to PROCRIT which could cause a rash over the shortness of breath, wheezing, reduction in fast pulse, or sweating.

of generalized allergy may be life-threatening. you are having a generalized allergic reaction, PROCRIT and notify a physician or emergency personnel immediately.

NOTES

a home dialysis patient and your doctor allows administer PROCRIT, please note the following: follow the instructions of your doctor concerning and administration of PROCRIT. Do not change instructions for administration of PROCRIT consulting your doctor.

will tell you what to do if you miss a dose of Always keep a spare syringe and needle on hand. consult your doctor if you notice anything unusual condition or your use of PROCRIT.

PREGNANCY

pregnant or nursing a baby, consult your physician PROCRIT.

by:
1080
Oaks, California 91320-1789
by:
Inc.
Jersey 08869-0670
U.S.A.

Revised May 1995

in Product Identification Guide, page 326

EMERGENCY telephone numbers, consult the Manufacturers Index.

Ortho Diagnostic Systems Inc.

A Johnson & Johnson Company
1001 U.S. HWY 202
RARITAN, NEW JERSEY 08869-0606

MICRhoGAM™ ℞

[*mike 'ro-gam*]
Rh_o (D) Immune Globulin (Human)
For Intramuscular Injection Only

Micro-Dose for use *only* after spontaneous or induced abortion or termination of ectopic pregnancy up to and including 12 weeks' gestation.

DESCRIPTION

MICRhoGAM Rh_o(D) Immune Globulin (Human) is a sterile solution containing IgG anti-Rh_o(D) for use in preventing Rh immunization in Rh negative individuals exposed to Rh positive red blood cells. A single dose of MICRhoGAM contains sufficient anti-Rh_o(D) (approximately 50 μg)† to suppress the immune response to 2.5 mL (or less) of Rh positive red blood cells.

All donors are carefully screened to eliminate those in high risk groups for disease transmission. Fractionation of the plasma is done by a modification of the cold alcohol procedure. Glycine (15 mg/mL) is included in the final product as a stabilizer. The final product contains 5% ± 1% globulin, 2.9 mg/mL sodium chloride, 0.01% polysorbate 80 and 0.003% thimerosal (mercury derivative).

This product is for intramuscular injection only.

† A full dose of Rh_o(D) Immune Globulin (Human) has traditionally been referred to as a "300 μg" dose and this usage is employed here for convenience in terminology. *It should not be construed as the actual anti-D content.* Each full dose of Rh_o(D) Immune Globulin (Human) must contain at least as much anti-D as 1 milliliter of the U.S. Reference Rh_o(D) Immune Globulin (Human). Studies performed at the Food and Drug Administration have shown that the U.S. Reference contains 820 international units (IU) of anti-D per milliliter. When the conversion factor determined for the International (WHO) Reference Preparation is used, 820 IU per milliliter is equivalent to 164 μg per milliliter of anti-D. MICRhoGAM contains approximately one-sixth the amount of anti-D contained in the full dose.

CLINICAL PHARMACOLOGY

Human immune globulins prepared by cold alcohol fractionation have not been reported to transmit hepatitis or other infectious diseases.

MICRhoGAM acts by suppressing the immune response of Rh negative women to Rh positive red blood cells. The risk of immunization is related to the number of Rh positive red cells received. The risk was found to be 3% when 0.1 mL of fetal red blood cells is present in the mother and 65% when 5 mL is present. In the first 12 weeks of gestation the total volume of red blood cells in the fetus is estimated at less than 2.5 mL.

Clinical studies demonstrated that administration of MICRhoGAM within three (3) hours following abortion was 100% effective in preventing Rh immunization. Studies in male volunteers showed MICRhoGAM to be effective when given as long as 72 hours after the infusion of Rh positive red cells. A lesser degree of protection is afforded if the antibody is administered beyond this time period.

INDICATIONS AND USAGE

MICRhoGAM is indicated for an Rh negative woman following spontaneous or induced abortion or termination of ectopic pregnancy up to and including 12 weeks' gestation, unless the father is conclusively shown to be Rh negative.

CONTRAINDICATIONS

MICRhoGAM must not be used for genetic amniocentesis at 15 to 18 weeks' gestation or antepartum prophylaxis at 28 weeks' gestation. RhoGAM™ Rh_o(D) Immune Globulin (Human) is recommended for any indication beyond 12 weeks' gestation.

Individuals known to have had an anaphylactic or severe systemic reaction to human globulin should not receive MICRhoGAM or any other Rh_o(D) Immune Globulin (Human).

WARNINGS

Do not inject intravenously.

PRECAUTIONS

Pregnancy Category C—Animal reproduction studies have not been conducted with MICRhoGAM. It is also not known whether Rh_o(D) Immune Globulin (Human) can cause fetal harm when administered to a pregnant woman or can affect reproduction capacity. Rh_o(D) Immune Globulin (Human) should be given to a pregnant woman only if clearly needed.

ADVERSE REACTIONS

Systemic reactions associated with administration of MICRhoGAM are extremely rare. Discomfort at the site of injection has been reported and a small number of women have noted a slight elevation in temperature.

DOSAGE AND ADMINISTRATION

Parenteral drug products should be inspected visually for particulate matter and discoloration prior to administration, whenever solution and container permit.

A single dose (approximately 50 μg)† of MICRhoGAM will completely suppress the immune response to 2.5 mL of Rh positive red blood cells (packed cells, not whole blood).

Administer a single dose of MICRhoGAM intramuscularly as soon as possible after termination of a pregnancy up to and including 12 weeks' gestation. At or beyond 13 weeks' gestation it is recommended that a single dose of RhoGAM Rh_o(D) Immune Globulin (Human) (approximately 300 μg)† be given instead of MICRhoGAM.

Since there is no lot number and expiration date on the prefilled syringes, they should not be removed from the protective pouch until immediately before use.

† See footnote under Description.

HOW SUPPLIED

—5 prefilled single-dose syringes of MICRhoGAM (Product code 780800) NDC 0562-8080-80
—package insert
—5 control forms
—5 patient identification cards
and
—25 prefilled single-dose syringes of MICRhoGAM (Product code 780820) NDC 0562-8080-82
—package insert
—25 control forms
—25 patient identification cards

STORAGE

Store at 2 to 8°C. DO NOT FREEZE.

RhoGAM™ ℞

[*ro 'gam*]
Rh_o (D) Immune Globulin (Human)
For Intramuscular Injection Only

DESCRIPTION

RhoGAM Rh_o(D) Immune Globulin (Human) is a sterile solution containing IgG anti-Rh_o(D) for use in preventing Rh immunization. Each single dose of RhoGAM contains sufficient anti-Rh_o(D) (approximately 300 μg)† to suppress the immune response to 15 mL (or less) of Rh positive red blood cells.

All donors are carefully screened to eliminate those in high risk groups for disease transmission. Fractionation of the plasma is done by a modification of the cold alcohol procedure. The final product contains 5% ± 1% globulin, 2.9 mg/mL sodium chloride, 0.01% polysorbate 80 and 0.003% thimerosal (mercury derivative), with glycine (15 mg/mL) as a stabilizer.

This product is for intramuscular injection only.

† A full dose of Rh_o(D) Immune Globulin (Human) has traditionally been referred to as a "300 μg" dose and this usage is employed here for convenience in terminology. *It should not be construed as the actual anti-D content.* Each full dose of Rh_o(D) Immune Globulin (Human) must contain at least as much anti-D as 1 milliliter of the U.S. Reference Rh_o(D) Immune Globulin (Human). Studies performed at the Food and Drug Administration have shown that the U.S. Reference contains 820 international units (IU) of anti-D per milliliter. When the conversion factor determined for the International (WHO) Reference Preparation is used, 820 IU per milliliter is equivalent to 164 μg per milliliter of anti-D.

CLINICAL PHARMACOLOGY

Human immune globulins prepared by cold alcohol fractionation have not been reported to transmit hepatitis or other infectious diseases.

RhoGAM acts by suppressing the immune response of Rh negative individuals to Rh positive red blood cells.

The obstetrical patient may be exposed to red blood cells from her Rh positive fetus during the normal course of pregnancy. Clinical studies proved that the incidence of Rh immunization as a result of pregnancy was reduced to 1% to 2% from 12% to 13% when RhoGAM was given within 72 hours following delivery. Further studies in which patients received Rh immune globulin, antepartum at 28 to 32 weeks and postpartum, reduced the risk of immunization to less than 0.1%.

An Rh negative individual transfused with one unit of Rh positive red blood cells has about an 80% likelihood of producing anti-Rh_o(D). Protection from Rh immunization is accomplished by administering the appropriate dose of RhoGAM.

INDICATIONS AND USAGE

Pregnancy and Other Obstetric Conditions

RhoGAM is indicated whenever it is known or suspected that fetal red cells have entered the circulation of an Rh negative

Continued on next page

PROCRIT®
EPOETIN ALFA

Full Prescribing Information

Manufactured by:
Amgen Inc.
One Amgen Center Drive
Thousand Oaks, CA 91320-1789

Distributed by:
Ortho Biotech Products, L.P.
Raritan, New Jersey 08869-0670

PROCRIT®
(Epoetin alfa)
FOR INJECTION

DESCRIPTION

Erythropoietin is a glycoprotein which stimulates red blood cell production. It is produced in the kidney and stimulates the division and differentiation of committed erythroid progenitors in the bone marrow. PROCRIT® (Epoetin alfa), a 165 amino acid glycoprotein manufactured by recombinant DNA technology, has the same biological effects as endogenous erythropoietin.[1] It has a molecular weight of 30,400 daltons and is produced by mammalian cells into which the human erythropoietin gene has been introduced. The product contains the identical amino acid sequence of isolated natural erythropoietin.

PROCRIT® is formulated as a sterile, colorless liquid in an isotonic sodium chloride/sodium citrate buffered solution or a sodium chloride/sodium phosphate buffered solution for intravenous (IV) or subcutaneous (SC) administration.

Single-dose, Preservative-free Vial: Each 1 mL of solution contains 2000, 3000, 4000 or 10,000 Units of Epoetin alfa, 2.5 mg Albumin (Human), 5.8 mg sodium citrate, 5.8 mg sodium chloride, and 0.06 mg citric acid in Water for Injection, USP (pH 6.9 ± 0.3). This formulation contains no preservative.

Single-dose, Preservative-free Vial: 1 mL (40,000 Units/mL). Each 1 mL of solution contains 40,000 Units of Epoetin alfa, 2.5 mg Albumin (Human), 1.2 mg sodium phosphate monobasic monohydrate, 1.8 mg sodium phosphate dibasic anhydrate, 0.7 mg sodium citrate, 5.8 mg sodium chloride, and 6.8 mcg citric acid in Water for Injection, USP (pH 6.9 ± 0.3). This formulation contains no preservative.

Multidose, Preserved Vial: 2 mL (20,000 Units, 10,000 Units/mL). Each 1 mL of solution contains 10,000 Units of Epoetin alfa, 2.5 mg Albumin (Human), 1.3 mg sodium citrate, 8.2 mg sodium chloride, 0.11 mg citric acid, and 1% benzyl alcohol as preservative in Water for Injection, USP (pH 6.1 ± 0.3).

Multidose, Preserved Vial: 1 mL (20,000 Units/mL). Each 1 mL of solution contains 20,000 Units of Epoetin alfa, 2.5 mg Albumin (Human), 1.3 mg sodium citrate, 8.2 mg sodium chloride, 0.11 mg citric acid, and 1% benzyl alcohol as preservative in Water for Injection, USP (pH 6.1 ± 0.3).

CLINICAL PHARMACOLOGY

Chronic Renal Failure Patients

Endogenous production of erythropoietin is normally regulated by the level of tissue oxygenation. Hypoxia and anemia generally increase the production of erythropoietin, which in turn stimulates erythropoiesis.[2] In normal subjects, plasma erythropoietin levels range from 0.01 to 0.03 Units/mL and increase up to 100- to 1000-fold during hypoxia or anemia.[2] In contrast, in patients with chronic renal failure (CRF), production of erythropoietin is impaired, and this erythropoietin deficiency is the primary cause of their anemia.[3,4]

Chronic renal failure is the clinical situation in which there is a progressive and usually irreversible decline in kidney function. Such patients may manifest the sequelae of renal dysfunction, including anemia, but do not necessarily require regular dialysis. Patients with end-stage renal disease (ESRD) are those patients with CRF who require regular dialysis or kidney transplantation for survival.

PROCRIT® has been shown to stimulate erythropoiesis in anemic patients with CRF, including both patients on dialysis and those who do not require regular dialysis.[4-13] The first evidence of a response to the three times weekly (TIW) administration of PROCRIT® is an increase in the reticulocyte count within 10 days, followed by increases in the red cell count, hemoglobin, and hematocrit, usually within 2 to 6 weeks.[4,5] Because of the length of time required for erythropoiesis – several days for erythroid progenitors to mature and be released into the circulation – a clinically significant increase in hematocrit is usually not observed in less than 2 weeks and may require up to 6 weeks in some patients. Once the hematocrit reaches the suggested target range (30% to 36%), that level can be sustained by PROCRIT® therapy in the absence of iron deficiency and concurrent illnesses.

The rate of hematocrit increase varies between patients and is dependent upon the dose of PROCRIT®, within a therapeutic range of approximately 50 to 300 Units/kg TIW.[4] A greater biologic response is not observed at doses exceeding 300 Units/kg TIW.[6] Other factors affecting the rate and extent of response include availability of iron stores, the baseline hematocrit, and the presence of concurrent medical problems.

Zidovudine-treated HIV-infected Patients

Responsiveness to PROCRIT® in HIV-infected patients is dependent upon the endogenous serum erythropoietin level prior to treatment. Patients with endogenous serum erythropoietin levels ≤ 500 mUnits/mL, and who are receiving a dose of zidovudine ≤ 4200 mg/week, may respond to PROCRIT® therapy. Patients with endogenous serum erythropoietin levels > 500 mUnits/mL do not appear to respond to PROCRIT® therapy. In a series of four clinical trials involving 255 patients, 60% to 80% of HIV-infected patients treated with zidovudine had endogenous serum erythropoietin levels ≤ 500 mUnits/mL.

Response to PROCRIT® in zidovudine-treated HIV-infected patients is manifested by reduced transfusion requirements and increased hematocrit.

Cancer Patients on Chemotherapy

A series of clinical trials enrolled 131 anemic cancer patients who received PROCRIT® TIW and who were receiving cyclic cisplatin- or non cisplatin-containing chemotherapy. Endogenous baseline serum erythropoietin levels varied among patients in these trials with approximately 75% (n = 83/110) having endogenous serum erythropoietin levels ≤ 132 mUnits/mL, and approximately 4% (n = 4/110) of patients having endogenous serum erythropoietin levels > 500 mUnits/mL. In general, patients with lower baseline serum erythropoietin levels responded more vigorously to PROCRIT® than patients with higher baseline erythropoietin levels. Although no specific serum erythropoietin level can be stipulated above which patients would be unlikely to respond to PROCRIT® therapy, treatment of patients with grossly elevated serum erythropoietin levels (eg, > 200 mUnits/mL) is not recommended.

Pharmacokinetics

In adult and pediatric patients with CRF, the elimination half-life of plasma erythropoietin after intravenously administered PROCRIT® ranges from 4 to 13 hours.[14-16] The half-life is

approximately 20% longer in CRF patients than that in healthy subjects. After SC administration, peak plasma levels are achieved within 5 to 24 hours. The half-life is similar between adult patients with serum creatinine level greater than 3 and not on dialysis and those maintained on dialysis. The pharmacokinetic data indicate no apparent difference in PROCRIT® half-life among adult patients above or below 65 years of age.

The pharmacokinetic profile of PROCRIT® in children and adolescents appears to be similar to that of adults. Limited data are available in neonates.[17] A study of 7 preterm very low birth weight neonates and 10 healthy adults given IV erythropoietin suggested that distribution volume was approximately 1.5 to 2 times higher in the preterm neonates than in the healthy adults, and clearance was approximately 3 times higher in the preterm neonates than in the healthy adults.[44]

The pharmacokinetics of PROCRIT® have not been studied in HIV-infected patients.

A pharmacokinetic study comparing 150 Units/kg SC TIW to 40,000 Units SC weekly dosing regimen was conducted for 4 weeks in healthy subjects (n = 12) and for 6 weeks in anemic cancer patients (n = 32) receiving cyclic chemotherapy. There was no accumulation of serum erythropoietin after the 2 dosing regimens during the study period. The 40,000 Units weekly regimen had a higher C_{max} (3- to 7-fold), longer T_{max} (2- to 3-fold), higher $AUC_{0\text{-}168h}$ (2- to 3-fold) of erythropoietin and lower clearance (50%) than the 150 Units/kg TIW regimen. In anemic cancer patients, the average $t_{1/2}$ was similar (40 hours with range of 16 to 67 hours) after both dosing regimens. After the 150 Units/kg TIW dosing, the values of T_{max} and clearance are similar (13.3 ± 12.4 vs. 14.2 ± 6.7 hours, and 20.2 ± 15.9 vs. 23.6 ± 9.5 mL/h/kg) between Week 1 when patients were receiving chemotherapy (n = 14) and Week 3 when patients were not receiving chemotherapy (n = 4). Differences were observed after the 40,000 Units weekly dosing with longer T_{max} (38 ± 18 hours) and lower clearance (9.2 ± 4.7 mL/h/kg) during Week 1 when patients were receiving chemotherapy (n = 18) compared with those (22 ± 4.5 hours, 13.9 ± 7.6 mL/h/kg) during Week 3 when patients were not receiving chemotherapy (n = 7).

The bioequivalence between the 10,000 Units/mL citrate-buffered Epoetin alfa formulation and the 40,000 Units/mL phosphate-buffered Epoetin alfa formulation has been demonstrated after SC administration of single 750 Units/kg doses to healthy subjects.

INDICATIONS AND USAGE

Treatment of Anemia of Chronic Renal Failure Patients

PROCRIT® is indicated for the treatment of anemia associated with CRF, including patients on dialysis (ESRD) and patients not on dialysis. PROCRIT® is indicated to elevate or maintain the red blood cell level (as manifested by the hematocrit or hemoglobin determinations) and to decrease the need for transfusions in these patients.

Non-dialysis patients with symptomatic anemia considered for therapy should have a hemoglobin less than 10 g/dL.

PROCRIT® is not intended for patients who require immediate correction of severe anemia. PROCRIT® may obviate the need for maintenance transfusions but is not a substitute for emergency transfusion.

Prior to initiation of therapy, the patient's iron stores should be evaluated. Transferrin saturation should be at least 20% and ferritin at least 100 ng/mL. Blood pressure should be adequately controlled prior to initiation of PROCRIT® therapy, and must be closely monitored and controlled during therapy.

PROCRIT® should be administered under the guidance of a qualified physician (see DOSAGE AND ADMINISTRATION).

Treatment of Anemia in Zidovudine-treated HIV-infected Patients

PROCRIT® is indicated for the treatment of anemia related to therapy with zidovudine in HIV-infected patients. PROCRIT® is indicated to elevate or maintain the red blood cell level (as manifested by the hematocrit or hemoglobin determinations) and to decrease the need for transfusions in these patients. PROCRIT® is not indicated for the treatment of anemia in HIV-infected patients due to other factors such as iron or folate deficiencies, hemolysis, or gastrointestinal bleeding, which should be managed appropriately.

PROCRIT®, at a dose of 100 Units/kg TIW, is effective in decreasing the transfusion requirement and increasing the red blood cell level of anemic, HIV-infected patients treated with zidovudine, when the endogenous serum erythropoietin level is ≤ 500 mUnits/mL and when patients are receiving a dose of zidovudine ≤ 4200 mg/week.

Treatment of Anemia in Cancer Patients on Chemotherapy

PROCRIT® is indicated for the treatment of anemia in patients with non-myeloid malignancies where anemia is due to the effect of concomitantly administered chemotherapy. PROCRIT® is indicated to decrease the need for transfusions in patients who will be receiving concomitant chemotherapy for a minimum of 2 months. PROCRIT® is not indicated for the treatment of anemia in cancer patients due to other factors such as iron or folate deficiencies, hemolysis, or gastrointestinal bleeding, which should be managed appropriately.

Reduction of Allogeneic Blood Transfusion in Surgery Patients

PROCRIT® is indicated for the treatment of anemic patients (hemoglobin > 10 to ≤ 13 g/dL) scheduled to undergo elective, noncardiac, nonvascular surgery to reduce the need for allogeneic blood transfusions.[18-20] PROCRIT® is indicated for patients at high risk for perioperative transfusions with significant, anticipated blood loss. PROCRIT® is not indicated for anemic patients who are willing to donate autologous blood. The safety of the perioperative use of PROCRIT® has been studied only in patients who are receiving anticoagulant prophylaxis.

CLINICAL EXPERIENCE: RESPONSE TO PROCRIT®

Chronic Renal Failure Patients

Response to PROCRIT® was consistent across all studies. In the presence of adequate iron stores (see IRON EVALUATION), the time to reach the target hematocrit is a function of the baseline hematocrit and the rate of hematocrit rise.

The rate of increase in hematocrit is dependent upon the dose of PROCRIT® administered and individual patient variation. In clinical trials at starting doses of 50 to 150 Units/kg TIW, adult patients responded with an average rate of hematocrit rise of:

Starting Dose (TIW IV)	Hematocrit Increase Points/Day	Points/2 Weeks
50 Units/kg	0.11	1.5
100 Units/kg	0.18	2.5
150 Units/kg	0.25	3.5

Over this dose range, approximately 95% of all patients responded with a clinically significant increase in hematocrit, and by the end of approximately 2 months of therapy virtually all patients were transfusion-independent. Changes in the quality of life of adult patients treated with PROCRIT® were assessed as part of a phase 3 clinical trial.[5,8] Once the target hematocrit (32% to 38%) was achieved, statistically significant improvements were demonstrated for most quality of life parameters measured, including energy and activity level, functional ability, sleep and eating behavior, health status, satisfaction with health, sex life, well-being, psychological effect, life satisfaction, and happiness. Patients also reported improvement in their disease symptoms. They showed a statistically significant increase in exercise capacity (VO_2 max), energy, and strength with a significant reduction in aching, dizziness, anxiety, shortness of breath, muscle weakness, and leg cramps.[8,21]

Adult Patients on Dialysis: Thirteen clinical studies were conducted, involving IV administration to a total of 1010 anemic patients on dialysis for 986 patient-years of PROCRIT® therapy. In the three largest of these clinical trials, the median maintenance dose necessary to maintain the hematocrit between 30% to 36% was approximately 75 Units/kg TIW. In the US multicenter phase 3 study, approximately 65% of the patients required doses of 100 Units/kg TIW, or less, to maintain their hematocrit at approximately 35%. Almost 10% of patients required a dose of 25 Units/kg, or less, and approximately 10% required a dose of more than 200 Units/kg TIW to maintain their hematocrit at this level.

A multicenter unit dose study was also conducted in 119 patients receiving peritoneal dialysis who self-administered PROCRIT® subcutaneously for approximately 109 patient-years of experience. Patients responded to PROCRIT® administered SC in a manner similar to patients receiving IV administration.[22]

Pediatric Patients on Dialysis: One hundred twenty-eight children from 2 months to 19 years of age with CRF requiring dialysis were enrolled in 4 clinical studies of PROCRIT®. The largest study was a placebo-controlled, randomized trial in 113 children with anemia (hematocrit ≤ 27%) undergoing peritoneal dialysis or hemodialysis. The initial dose of PROCRIT® was 50 Units/kg IV or SC TIW. The dose of study drug was titrated to achieve either a hematocrit of 30% to 36% or an absolute increase in hematocrit of 6 percentage points over baseline.

At the end of the initial 12 weeks, a statistically significant rise in mean hematocrit (9.4% vs 0.9%) was observed only in the PROCRIT® arm. The proportion of children achieving a hematocrit of 30%, or an increase in hematocrit of 6 percentage points over baseline, at any time during the first 12 weeks was higher in the PROCRIT® arm (96% vs 58%). Within 12 weeks of initiating PROCRIT® therapy, 92.3% of the pediatric patients were transfusion-independent as compared to 65.4% who received placebo. Among patients who received 36 weeks of PROCRIT®, hemodialysis

patients required a higher median maintenance dose (167 Units/kg/week [n = 28] vs 76 Units/kg/week [n = 36]) and took longer to achieve a hematocrit of 30% to 36% (median time to response 69 days vs 32 days) than patients undergoing peritoneal dialysis.

Patients With CRF Not Requiring Dialysis

Four clinical trials were conducted in patients with CRF not on dialysis involving 181 patients treated with PROCRIT® for approximately 67 patient-years of experience. These patients responded to PROCRIT® therapy in a manner similar to that observed in patients on dialysis. Patients with CRF not on dialysis demonstrated a dose-dependent and sustained increase in hematocrit when PROCRIT® was administered by either an IV or SC route, with similar rates of rise of hematocrit when PROCRIT® was administered by either route. Moreover, PROCRIT® doses of 75 to 150 Units/kg per week have been shown to maintain hematocrits of 36% to 38% for up to 6 months. Correcting the anemia of progressive renal failure will allow patients to remain active even though their renal function continues to decrease.[23-24]

Zidovudine-treated HIV-infected Patients

PROCRIT® has been studied in four placebo-controlled trials enrolling 297 anemic (hematocrit < 30%) HIV-infected (AIDS) patients receiving concomitant therapy with zidovudine (all patients were treated with Epoetin alfa manufactured by Amgen Inc). In the subgroup of patients (89/125 PROCRIT® and 88/130 placebo) with prestudy endogenous serum erythropoietin levels ≤ 500 mUnits/mL, PROCRIT® reduced the mean cumulative number of units of blood transfused per patient by approximately 40% as compared to the placebo group.[25] Among those patients who required transfusions at baseline, 43% of patients treated with PROCRIT® versus 18% of placebo-treated patients were transfusion-independent during the second and third months of therapy. PROCRIT® therapy also resulted in significant increases in hematocrit in comparison to placebo. When examining the results according to the weekly dose of zidovudine received during month 3 of therapy, there was a statistically significant ($p < 0.003$) reduction in transfusion requirements in patients treated with PROCRIT® (n = 51) compared to placebo treated patients (n = 54) whose mean weekly zidovudine dose was ≤ 4200 mg/week.[25]

Approximately 17% of the patients with endogenous serum erythropoietin levels ≤ 500 mUnits/mL receiving PROCRIT® in doses from 100 to 200 Units/kg TIW achieved a hematocrit of 38% without administration of transfusions or significant reduction in zidovudine dose. In the subgroup of patients whose prestudy endogenous serum erythropoietin levels were > 500 mUnits/mL, PROCRIT® therapy did not reduce transfusion requirements or increase hematocrit, compared to the corresponding responses in placebo-treated patients.

In a 6 month open-label PROCRIT® study, patients responded with decreased transfusion requirements and sustained increases in hematocrit and hemoglobin with doses of PROCRIT® up to 300 Units/kg TIW.[25-27]

Responsiveness to PROCRIT® therapy may be blunted by intercurrent infectious/inflammatory episodes and by an increase in zidovudine dosage. Consequently, the dose of PROCRIT® must be titrated based on these factors to maintain the desired erythropoietic response.

Cancer Patients on Chemotherapy

Three-Times Weekly (TIW) Dosing

PROCRIT® administered TIW has been studied in a series of six placebo-controlled, double-blind trials that enrolled 131 anemic cancer patients receiving PROCRIT® or matching placebo. Across all studies, 72 patients were treated with concomitant non cisplatin-containing chemotherapy regimens and 59 patients were treated with concomitant cisplatin-containing chemotherapy regimens. Patients were randomized to PROCRIT® 150 Units/kg or placebo subcutaneously TIW for 12 weeks in each study.

The results of the pooled data from these six studies are shown in the table below. Because of the length of time required for erythropoiesis and red cell maturation, the efficacy of PROCRIT® (reduction in proportion of patients requiring transfusions) is not manifested until 2 to 6 weeks after initiation of PROCRIT®.

Proportion of Patients Transfused During Chemotherapy (Efficacy Population[a])

Chemotherapy Regimen	On Study[b]		During Months 2 and 3[c]	
	PROCRIT®	Placebo	PROCRIT®	Placebo
Regimens without cisplatin	44% (15/34)	44% (16/36)	21% (6/29)	33% (11/33)
Regimens containing cisplatin	50% (14/28)	63% (19/30)	23% (5/22)[d]	56% (14/25)
Combined	47% (29/62)	53% (35/66)	22% (11/51)[d]	43% (25/58)

[a] Limited to patients remaining on study at least 15 days (1 patient excluded from PROCRIT®, 2 patients excluded from placebo).

[b] Includes all transfusions from day 1 through the end of study.

[c] Limited to patients remaining on study beyond week 6 and includes only transfusions during weeks 5-12.

[d] Unadjusted 2-sided $p < 0.05$.

Intensity of chemotherapy in the above trials was not directly assessed, however the degree and timing of neutropenia was comparable across all trials. Available evidence suggests that patients with lymphoid and solid cancers respond similarly to PROCRIT® therapy, and that patients with or without tumor infiltration of the bone marrow respond similarly to PROCRIT® therapy.

Weekly (QW) Dosing

PROCRIT® was also studied in a placebo-controlled, double-blind trial utilizing weekly dosing in a total of 344 anemic cancer patients. In this trial, 61 (35 placebo arm and 26 in the PROCRIT® arm) patients were treated with concomitant cisplatin containing regimens and 283 patients received concomitant chemotherapy regimens that did not contain cisplatinum. Patients were randomized to PROCRIT® 40,000 Units weekly (n = 174) or placebo (n = 170) SC for a planned treatment period of 16 weeks. If hemoglobin had not increased by > 1 g/dL, after 4 weeks of therapy or the patient received RBC transfusion during the first 4 weeks of therapy, study drug was increased to 60,000 Units weekly. Forty-three percent of patients in the Epoetin alfa group required an increase in PROCRIT® dose to 60,000 Units weekly.[25]

Results demonstrated that PROCRIT® therapy reduced the proportion of patients transfused in day 29 through week 16

of the study as compared to placebo. Twenty-five patients (14%) in the PROCRIT® group received transfusions compared to 48 patients (28%) in the placebo group ($p = 0.0010$) between day 29 and week 16 or the last day on study.

Comparable intensity of chemotherapy for patients enrolled in the two study arms was suggested by similarities in mean dose and frequency of administration for the 10 most commonly administered chemotherapy agents, and similarity in the incidence of changes in chemotherapy during the trial in the two arms.

Surgery Patients

PROCRIT® has been studied in a placebo-controlled, double-blind trial enrolling 316 patients scheduled for major, elective orthopedic hip or knee surgery who were expected to require ≥ 2 units of blood and who were not able or willing to participate in an autologous blood donation program. Based on previous studies which demonstrated that pretreatment hemoglobin is a predictor of risk of receiving transfusion,[20,28] patients were stratified into one of three groups based on their pretreatment hemoglobin [≤ 10 (n = 2), > 10 to ≤ 13 (n = 96), and > 13 to ≤ 15 g/dL (n = 218)] and then randomly assigned to receive 300 Units/kg PROCRIT®, 100 Units/kg PROCRIT® or placebo by SC injection for 10 days before surgery, on the day of surgery, and for 4 days after surgery.[18] All patients received oral iron and a low-dose post-operative warfarin regimen.[18]

Treatment with PROCRIT® 300 Units/kg significantly ($p = 0.024$) reduced the risk of allogeneic transfusion in patients with a pretreatment hemoglobin of > 10 to ≤ 13; 5/31 (16%) of PROCRIT® 300 Units/kg, 6/26 (23%) of PROCRIT® 100 Units/kg, and 13/29 (45%) of placebo-treated patients were transfused.[18] There was no significant difference in the number of patients transfused between PROCRIT® (9% 300 Units/kg, 6% 100 Units/kg) and placebo (13%) in the > 13 to ≤ 15 g/dL hemoglobin stratum. There were too few patients in the ≤ 10 g/dL group to determine if PROCRIT® is useful in this hemoglobin strata. In the > 10 to ≤ 13 g/dL pretreatment stratum, the mean number of units transfused per PROCRIT®-treated patient (0.45 units blood for 300 Units/kg, 0.42 units blood for 100 Units/kg) was less than the mean transfused per placebo-treated patient (1.14 units) (overall $p = 0.028$). In addition, mean hemoglobin, hematocrit, and reticulocyte counts increased significantly during the presurgery period in patients treated with PROCRIT®.[18]

PROCRIT® was also studied in an open-label, parallel-group trial enrolling 145 subjects with a pretreatment hemoglobin level of ≥ 10 to ≤ 13 g/dL who were scheduled for major orthopedic hip or knee surgery and who were not participating in an autologous program.[19] Subjects were randomly assigned to receive one of two SC dosing regimens of PROCRIT® (600 Units/kg once weekly for 3 weeks prior to surgery and on the day of surgery, or 300 Units/kg once daily for 10 days prior to surgery, on the day of surgery and for 4 days after surgery). All subjects received oral iron and appropriate pharmacologic anticoagulation therapy.

From pretreatment to presurgery, the mean increase in hemoglobin in the 600 Units/kg weekly group (1.44 g/dL) was greater than observed in the 300 Units/kg daily group.[19] The mean increase in absolute reticulocyte count was smaller in the weekly group ($0.11 \times 10^6/mm^3$) compared to

the daily group (0.17 x 10^6/mm^3). Mean hemoglobin levels were similar for the two treatment groups throughout the postsurgical period.

The erythropoietic response observed in both treatment groups resulted in similar transfusion rates [11/69 (16%) in the 600 Units/kg weekly group and 14/71 (20%) in the 300 Units/kg daily group].[19] The mean number of units transfused per subject was approximately 0.3 units in both treatment groups.

CONTRAINDICATIONS

PROCRIT® is contraindicated in patients with:

1. Uncontrolled hypertension.
2. Known hypersensitivity to mammalian cell-derived products.
3. Known hypersensitivity to Albumin (Human).

WARNINGS

Pediatric Use

The multidose preserved formulation contains benzyl alcohol. Benzyl alcohol has been reported to be associated with an increased incidence of neurological and other complications in premature infants which are sometimes fatal.

Thrombotic Events and Increased Mortality

A randomized, prospective trial of 1265 hemodialysis patients with clinically evident cardiac disease (ischemic heart disease or congestive heart failure) was conducted in which patients were assigned to PROCRIT® treatment targeted to a maintenance hematocrit of either 42 ± 3% or 30 ± 3%.[42] Increased mortality was observed in 634 patients randomized to a target hematocrit of 42% [221 deaths (35% mortality)] compared to 631 patients targeted to remain at a hematocrit of 30% [185 deaths (29% mortality)]. The reason for the increased mortality observed in these studies is unknown, however, the incidence of non-fatal myocardial infarctions (3.1% vs 2.3%), vascular access thromboses (39% vs 29%), and all other thrombotic events (22% vs 18%) were also higher in the group randomized to achieve a hematocrit of 42%.

Increased mortality was also observed in a randomized placebo-controlled study of PROCRIT® in adult patients who did not have CRF who were undergoing coronary artery bypass surgery (7 deaths in 126 patients randomized to PROCRIT® versus no deaths among 56 patients receiving placebo). Four of these deaths occurred during the period of study drug administration and all four deaths were associated with thrombotic events. While the extent of the population affected is unknown, in patients at risk for thrombosis, the anticipated benefits of PROCRIT® treatment should be weighed against the potential for increased risks associated with therapy.

In a randomized, prospective trial conducted with another Epoetin alfa product, in 939 women with metastatic carcinoma of the breast who were receiving chemotherapy, patients were assigned to receive either Epoetin alfa or placebo for up to a year, in a weekly schedule, with the primary goal of showing improved survival and improved quality of life in the Epoetin alfa treatment arm.[25] This study utilized a treatment strategy designed to maintain hemoglobin levels of 12 to 14 g/dL (hematocrit 36 to 42%). Increased mortality in the first 4 months after randomization was observed among 469 patients who received the erythropoietin product [41 deaths (8.7% mortality)] compared to 470 patients who received

placebo [16 deaths (3.4% mortality)]. In the first four months of the study, the incidence of fatal thrombotic vascular events (1.1% vs 0.2%) and death attributed to disease progression (6.0% vs 2.8%) were both higher in the group randomized to receive Epoetin alfa as compared to placebo. Based on Kaplan-Meier estimates, the proportion of subjects surviving at 12 months after randomization was lower in the Epoetin alfa group than in the placebo group (70% vs 76%), $p = 0.012$, log rank. However, due to insufficient monitoring and data collection, reliable comparisons cannot be made concerning the effect of Epoetin alfa on overall time to disease progression, progression-free survival, and overall survival.

Pure Red Cell Aplasia

Pure red cell aplasia (PRCA), in association with neutralizing antibodies to native erythropoietin, has been observed in patients treated with recombinant erythropoietins. PRCA has been reported in a limited number of patients exposed to PROCRIT®. This has been reported predominantly in patients with CRF. Any patient with loss of response to PROCRIT® should be evaluated for the etiology of loss of effect (see PRECAUTIONS: LACK OR LOSS OF RESPONSE). PROCRIT® should be discontinued in any patient with evidence of PRCA and the patient evaluated for the presence of binding and neutralizing antibodies to PROCRIT®, native erythropoietin, and any other recombinant erythropoietin administered to the patient. Amgen/Ortho Biotech Products, L.P. should be contacted to assist in this evaluation. In patients with PRCA secondary to neutralizing antibodies to erythropoietin, PROCRIT® should not be administered and such patients should not be switched to another product as anti-erythropoietin antibodies cross-react with other erythropoietins (see ADVERSE REACTIONS).

Albumin (Human)

PROCRIT® contains albumin, a derivative of human blood. Based on effective donor screening and product manufacturing processes, it carries an extremely remote risk for transmission of viral diseases. A theoretical risk for transmission of Creutzfeldt-Jakob disease (CJD) also is considered extremely remote. No cases of transmission of viral diseases or CJD have ever been identified for albumin.

Chronic Renal Failure Patients

Hypertension: Patients with uncontrolled hypertension should not be treated with PROCRIT®; blood pressure should be controlled adequately before initiation of therapy. Up to 80% of patients with CRF have a history of hypertension.[29] Although there does not appear to be any direct pressor effects of PROCRIT®, blood pressure may rise during PROCRIT® therapy. During the early phase of treatment when the hematocrit is increasing, approximately 25% of patients on dialysis may require initiation of, or increases in, antihypertensive therapy. Hypertensive encephalopathy and seizures have been observed in patients with CRF treated with PROCRIT®.

Special care should be taken to closely monitor and aggressively control blood pressure in patients treated with PROCRIT®. Patients should be advised as to the importance of compliance with antihypertensive therapy and dietary restrictions. If blood pressure is difficult to control by initiation of appropriate measures, the hemoglobin may be reduced by decreasing or withholding the dose of PROCRIT®. A clinically significant decrease in hemoglobin may not be observed for several weeks.

It is recommended that the dose of PROCRIT® be decreased if the hemoglobin increase exceeds 1 g/dL in any 2-week period, because of the possible association of excessive rate of rise of hemoglobin with an exacerbation of hypertension. In CRF patients on hemodialysis with clinically evident ischemic heart disease or congestive heart failure, the hemoglobin should be managed carefully, not to exceed 12 g/dL (see THROMBOTIC EVENTS).

Seizures: Seizures have occurred in patients with CRF participating in PROCRIT® clinical trials.

In adult patients on dialysis, there was a higher incidence of seizures during the first 90 days of therapy (occurring in approximately 2.5% of patients) as compared with later timepoints.

Given the potential for an increased risk of seizures during the first 90 days of therapy, blood pressure and the presence of premonitory neurologic symptoms should be monitored closely. Patients should be cautioned to avoid potentially hazardous activities such as driving or operating heavy machinery during this period.

While the relationship between seizures and the rate of rise of hemoglobin is uncertain, it is recommended that the dose of PROCRIT® be decreased if the hemoglobin increase exceeds 1 g/dL in any 2-week period.

Thrombotic Events: During hemodialysis, patients treated with PROCRIT® may require increased anticoagulation with heparin to prevent clotting of the artificial kidney (see ADVERSE REACTIONS for more information about thrombotic events).

Other thrombotic events (eg, myocardial infarction, cerebrovascular accident, transient ischemic attack) have occurred in clinical trials at an annualized rate of less than 0.04 events per patient year of PROCRIT® therapy. These trials were conducted in adult patients with CRF (whether on dialysis or not) in whom the target hematocrit was 32% to 40%. However, the risk of thrombotic events, including vascular access thrombosis, was significantly increased in adult patients with ischemic heart disease or congestive heart failure receiving PROCRIT® therapy with the goal of reaching a normal hematocrit (42%) as compared to a target hematocrit of 30%. Patients with pre-existing cardiovascular disease should be monitored closely.

Zidovudine-treated HIV-infected Patients

In contrast to CRF patients, PROCRIT® therapy has not been linked to exacerbation of hypertension, seizures, and thrombotic events in HIV-infected patients.

PRECAUTIONS

The parenteral administration of any biologic product should be attended by appropriate precautions in case allergic or other untoward reactions occur (see CONTRAINDICATIONS). In clinical trials, while transient rashes were occasionally observed concurrently with PROCRIT® therapy, no serious allergic or anaphylactic reactions were reported (see ADVERSE REACTIONS for more information regarding allergic reactions).

The safety and efficacy of PROCRIT® therapy have not been established in patients with a known history of a seizure disorder or underlying hematologic disease (eg, sickle cell anemia, myelodysplastic syndromes, or hypercoagulable disorders).

In some female patients, menses have resumed following PROCRIT® therapy; the possibility of pregnancy should be discussed and the need for contraception evaluated.

Hematology

Exacerbation of porphyria has been observed rarely in patients with CRF treated with PROCRIT®. However, PROCRIT® has not caused increased urinary excretion of porphyrin metabolites in normal volunteers, even in the presence of a rapid erythropoietic response. Nevertheless, PROCRIT® should be used with caution in patients with known porphyria.

In preclinical studies in dogs and rats, but not in monkeys, PROCRIT® therapy was associated with subclinical bone marrow fibrosis. Bone marrow fibrosis is a known complication of CRF in humans and may be related to secondary hyperparathyroidism or unknown factors. The incidence of bone marrow fibrosis was not increased in a study of adult patients on dialysis who were treated with PROCRIT® for 12 to 19 months, compared to the incidence of bone marrow fibrosis in a matched group of patients who had not been treated with PROCRIT®.

Hemoglobin in CRF patients should be measured twice a week; zidovudine-treated HIV-infected and cancer patients should have hemoglobin measured once a week until hemoglobin has been stabilized, and measured periodically thereafter.

Lack or Loss of Response

If the patient fails to respond or to maintain a response to doses within the recommended dosing range, the following etiologies should be considered and evaluated:

1. Iron deficiency: Virtually all patients will eventually require supplemental iron therapy (see IRON EVALUATION).
2. Underlying infectious, inflammatory, or malignant processes.
3. Occult blood loss.
4. Underlying hematologic diseases (ie, thalassemia, refractory anemia, or other myelodysplastic disorders).
5. Vitamin deficiencies: Folic acid or vitamin B12.
6. Hemolysis.
7. Aluminum intoxication.
8. Osteitis fibrosa cystica.
9. Pure Red Cell Aplasia (PRCA): In the absence of another etiology, the patient should be evaluated for evidence of PRCA and sera should be tested for the presence of antibodies to recombinant erythropoietins.

Iron Evaluation

During PROCRIT® therapy, absolute or functional iron deficiency may develop. Functional iron deficiency, with normal ferritin levels but low transferrin saturation, is presumably due to the inability to mobilize iron stores rapidly enough to support increased erythropoiesis. Transferrin saturation should be at least 20% and ferritin should be at least 100 ng/mL.

Prior to and during PROCRIT® therapy, the patient's iron status, including transferrin saturation (serum iron divided by iron binding capacity) and serum ferritin, should be evaluated. Virtually all patients will eventually require supplemental iron to increase or maintain transferrin saturation to levels which will adequately support erythropoiesis stimulated by PROCRIT®. All surgery patients being treated with PROCRIT®

should receive adequate iron supplementation throughout the course of therapy in order to support erythropoiesis and avoid depletion of iron stores.

Drug Interactions

No evidence of interaction of PROCRIT® with other drugs was observed in the course of clinical trials.

Carcinogenesis, Mutagenesis, and Impairment of Fertility

Carcinogenic potential of PROCRIT® has not been evaluated. PROCRIT® does not induce bacterial gene mutation (Ames Test), chromosomal aberrations in mammalian cells, micronuclei in mice, or gene mutation at the HGPRT locus. In female rats treated IV with PROCRIT®, there was a trend for slightly increased fetal wastage at doses of 100 and 500 Units/kg.

Pregnancy Category C

PROCRIT® has been shown to have adverse effects in rats when given in doses 5 times the human dose. There are no adequate and well-controlled studies in pregnant women. PROCRIT® should be used during pregnancy only if potential benefit justifies the potential risk to the fetus.

In studies in female rats, there were decreases in body weight gain, delays in appearance of abdominal hair, delayed eyelid opening, delayed ossification, and decreases in the number of caudal vertebrae in the F1 fetuses of the 500 Units/kg group. In female rats treated IV, there was a trend for slightly increased fetal wastage at doses of 100 and 500 Units/kg. PROCRIT® has not shown any adverse effect at doses as high as 500 Units/kg in pregnant rabbits (from day 6 to 18 of gestation).

Nursing Mothers

Postnatal observations of the live offspring (F1 generation) of female rats treated with PROCRIT® during gestation and lactation revealed no effect of PROCRIT® at doses of up to 500 Units/kg. There were, however, decreases in body weight gain, delays in appearance of abdominal hair, eyelid opening, and decreases in the number of caudal vertebrae in the F1 fetuses of the 500 Units/kg group. There were no PROCRIT®-related effects on the F2 generation fetuses.

It is not known whether PROCRIT® is excreted in human milk. Because many drugs are excreted in human milk, caution should be exercised when PROCRIT® is administered to a nursing woman.

Pediatric Use

See WARNINGS: PEDIATRIC USE.

Pediatric Patients on Dialysis: PROCRIT® is indicated in infants (1 month to 2 years), children (2 years to 12 years), and adolescents (12 years to 16 years) for the treatment of anemia associated with CRF requiring dialysis. Safety and effectiveness in pediatric patients less than 1 month old have not been established (see CLINICAL EXPERIENCE: CHRONIC RENAL FAILURE, PEDIATRIC PATIENTS ON DIALYSIS). The safety data from these studies show that there is no increased risk to pediatric CRF patients on dialysis when compared to the safety profile of PROCRIT® in adult CRF patients (see ADVERSE REACTIONS and WARNINGS). Published literature[30-33] provides supportive evidence of the safety and effectiveness of PROCRIT® in pediatric CRF patients on dialysis.

Pediatric Patients Not Requiring Dialysis: Published literature[33,34] has reported the use of PROCRIT® in 133 pediatric patients with anemia associated with CRF not requiring dialysis, ages 3 months to 20 years, treated with 50 to 250 Units/kg SC or IV, QW to TIW. Dose-dependent increases in hemoglobin and hematocrit were observed with reductions in transfusion requirements.

Pediatric HIV-infected Patients: Published literature[35,36] has reported the use of PROCRIT® in 20 zidovudine-treated anemic HIV-infected pediatric patients ages 8 months to 17 years, treated with 50 to 400 Units/kg SC or IV, 2 to 3 times per week. Increases in hemoglobin levels and in reticulocyte counts, and decreases in or elimination of blood transfusions were observed.

Pediatric Cancer Patients on Chemotherapy: Published literature[37,38] has reported the use of PROCRIT® in approximately 64 anemic pediatric cancer patients ages 6 months to 18 years, treated with 25 to 300 Units/kg SC or IV, 3 to 7 times per week. Increases in hemoglobin and decreases in transfusion requirements were noted.

Geriatric Use

Among 1051 patients enrolled in the 5 clinical trials of PROCRIT® for reduction of allogeneic blood transfusions in patients undergoing elective surgery 745 received PROCRIT® and 306 received placebo. Of the 745 patients who received PROCRIT®, 432 (58%) were aged 65 and over, while 175 (23%) were 75 and over. No overall differences in safety or effectiveness were observed between geriatric and younger patients. The dose requirements for PROCRIT® in geriatric and younger patients within the 4 trials using the TIW schedule were similar. Insufficient numbers of patients were enrolled in the study using the weekly dosing regimen to determine whether the dosing requirements differ for this schedule.

Of the 882 patients enrolled in the 3 studies of chronic renal failure patients on dialysis, 757 received PROCRIT® and 125 received placebo. Of the 757 patients who received PROCRIT®, 361 (47%) were aged 65 and over, while 100 (13%) were 75 and over. No differences in safety or effectiveness were observed between geriatric and younger patients. Dose selection and adjustment for an elderly patient should be individualized to achieve and maintain the target hematocrit (See DOSAGE AND ADMINISTRATION).

Insufficient numbers of patients age 65 or older were enrolled in clinical studies of PROCRIT® for the treatment of anemia associated with pre-dialysis chronic renal failure, cancer chemotherapy, and Zidovudine-treatment of HIV infection to determine whether they respond differently from younger subjects.

Chronic Renal Failure Patients

Patients with CRF Not Requiring Dialysis

Blood pressure and hemoglobin should be monitored no less frequently than for patients maintained on dialysis. Renal function and fluid and electrolyte balance should be closely monitored, as an improved sense of well-being may obscure the need to initiate dialysis in some patients.

Hematology

Sufficient time should be allowed to determine a patient's responsiveness to a dosage of PROCRIT® before adjusting

the dose. Because of the time required for erythropoiesis and the red cell half-life, an interval of 2 to 6 weeks may occur between the time of a dose adjustment (initiation, increase, decrease, or discontinuation) and a significant change in hemoglobin.

In order to avoid reaching the suggested target hemoglobin too rapidly, or exceeding the suggested target range (hemoglobin of 10 g/dL to 12 g/dL), the guidelines for dose and frequency of dose adjustments (see DOSAGE AND ADMINISTRATION) should be followed.

For patients who respond to PROCRIT® with a rapid increase in hemoglobin (eg, more than 1 g/dL in any 2-week period), the dose of PROCRIT® should be reduced because of the possible association of excessive rate of rise of hemoglobin with an exacerbation of hypertension.

The elevated bleeding time characteristic of CRF decreases toward normal after correction of anemia in adult patients treated with PROCRIT®. Reduction of bleeding time also occurs after correction of anemia by transfusion.

Laboratory Monitoring

The hemoglobin should be determined twice a week until it has stabilized in the suggested target range and the maintenance dose has been established. After any dose adjustment, the hemoglobin should also be determined twice weekly for at least 2 to 6 weeks until it has been determined that the hemoglobin has stabilized in response to the dose change. The hemoglobin should then be monitored at regular intervals.

A complete blood count with differential and platelet count should be performed regularly. During clinical trials, modest increases were seen in platelets and white blood cell counts. While these changes were statistically significant, they were not clinically significant and the values remained within normal ranges.

In patients with CRF, serum chemistry values (including blood urea nitrogen [BUN], uric acid, creatinine, phosphorus, and potassium) should be monitored regularly. During clinical trials in adult patients on dialysis, modest increases were seen in BUN, creatinine, phosphorus, and potassium. In some adult patients with CRF not on dialysis treated with PROCRIT®, modest increases in serum uric acid and phosphorus were observed. While changes were statistically significant, the values remained within the ranges normally seen in patients with CRF.

Diet

As the hemoglobin increases and patients experience an improved sense of well-being and quality of life, the importance of compliance with dietary and dialysis prescriptions should be reinforced. In particular, hyperkalemia is not uncommon in patients with CRF. In US studies in patients on dialysis, hyperkalemia has occurred at an annualized rate of approximately 0.11 episodes per patient-year of PROCRIT® therapy, often in association with poor compliance to medication, diet, and/or dialysis.

Dialysis Management

Therapy with PROCRIT® results in an increase in hematocrit and a decrease in plasma volume which could affect dialysis efficiency. In studies to date, the resulting increase in hematocrit did not appear to adversely affect dialyzer

function[9,10] or the efficiency of high flux hemodialysis.[11] During hemodialysis, patients treated with PROCRIT® may require increased anticoagulation with heparin to prevent clotting of the artificial kidney.

Patients who are marginally dialyzed may require adjustments in their dialysis prescription. As with all patients on dialysis, the serum chemistry values (including BUN, creatinine, phosphorus, and potassium) in patients treated with PROCRIT® should be monitored regularly to assure the adequacy of the dialysis prescription.

Information for Patients

In those situations in which the physician determines that a home dialysis patient can safely and effectively self-administer PROCRIT®, the patient should be instructed as to the proper dosage and administration. Home dialysis patients should be referred to the full "Information For Home Dialysis Patients" insert; it is not a disclosure of all possible effects. Patients should be informed of the signs and symptoms of allergic drug reaction and advised of appropriate actions. If home use is prescribed for a home dialysis patient, the patient should be thoroughly instructed in the importance of proper disposal and cautioned against the reuse of needles, syringes, or drug product. A puncture-resistant container for the disposal of used syringes and needles should be available to the patient. The full container should be disposed of according to the directions provided by the physician.

Renal Function

In adult patients with CRF not on dialysis, renal function and fluid and electrolyte balance should be closely monitored, as an improved sense of well-being may obscure the need to initiate dialysis in some patients. In patients with CRF not on dialysis, placebo-controlled studies of progression of renal dysfunction over periods of greater than 1 year have not been completed. In shorter term trials in adult patients with CRF not on dialysis, changes in creatinine and creatinine clearance were not significantly different in patients treated with PROCRIT® compared with placebo-treated patients. Analysis of the slope of 1/serum creatinine versus time plots in these patients indicates no significant change in the slope after the initiation of PROCRIT® therapy.

Zidovudine-treated HIV-infected Patients

Hypertension

Exacerbation of hypertension has not been observed in zidovudine-treated HIV-infected patients treated with PROCRIT®. However, PROCRIT® should be withheld in these patients if pre-existing hypertension is uncontrolled, and should not be started until blood pressure is controlled. In double-blind studies, a single seizure has been experienced by a patient treated with PROCRIT®.[25]

Cancer Patients on Chemotherapy

Hypertension

Hypertension, associated with a significant increase in hemoglobin, has been noted rarely in patients treated with PROCRIT®. Nevertheless, blood pressure in patients treated with PROCRIT® should be monitored carefully, particularly in patients with an underlying history of hypertension or cardiovascular disease.

Seizures

In double-blind, placebo-controlled trials, 3.2% (n = 2/63) of patients treated with PROCRIT® TIW and 2.9% (n = 2/68) of

placebo-treated patients had seizures. Seizures in 1.6% (n = 1/63) of patients treated with PROCRIT® TIW occurred in the context of a significant increase in blood pressure and hematocrit from baseline values. However, both patients treated with PROCRIT® also had underlying CNS pathology which may have been related to seizure activity.

In a placebo-controlled, double-blind trial utilizing weekly dosing with PROCRIT®, 1.2% (n = 2/168) of safety-evaluable patients treated with PROCRIT® and 1% (n = 1/165) of placebo-treated patients had seizures. Seizures in the patients treated with weekly PROCRIT® occurred in the context of a significant increase in hemoglobin from baseline values however significant increases in blood pressure were not seen. These patients may have had other CNS pathology.

Thrombotic Events

In double-blind, placebo-controlled trials, 3.2% (n = 2/63) of patients treated with PROCRIT® TIW and 11.8% (n = 8/68) of placebo-treated patients had thrombotic events (eg, pulmonary embolism, cerebrovascular accident) (See WARNINGS; Thrombotic Events and Increased Mortality).

In a placebo-controlled, double-blind trial utilizing weekly dosing with PROCRIT®, 6.0% (n = 10/168) of safety-evaluable patients treated with PROCRIT® and 3.6% (n = 6/165) (p = 0.444) of placebo-treated patients had clinically significant thrombotic events (deep vein thrombosis requiring anticoagulant therapy, embolic event including pulmonary embolism, myocardial infarction, cerebral ischemia, left ventricular failure and thrombotic micro-angiopathy). A definitive relationship between the rate of hemoglobin increase and the occurrence of clinically significant thrombotic events could not be evaluated due to the limited schedule of hemoglobin measurements in this study.

Tumor Growth Factor Potential

PROCRIT® is a growth factor that primarily stimulates red cell production. Erythropoietin receptors are also found to be present on the surface of some malignant cell lines and tumor biopsy specimens. However, it is not known if these receptors are functional. A randomized, placebo-controlled trial was conducted in 224 chemotherapy-naïve, non-anemic patients with small cell lung cancer receiving cisplatin-based combination chemotherapy, to investigate whether the concurrent use of PROCRIT® stimulated tumor growth as assessed by impact on overall response rate. Patients were randomized to receive PROCRIT® 150 Units/kg or placebo subcutaneously TIW during chemotherapy. The overall response rates, after 3 cycles of treatment, were 72% and 67%, in the PROCRIT® and placebo arms, respectively. Complete response rates (17% vs. 14%) and median overall survival (10.5 mos vs. 10.4 mos) were similar in the PROCRIT® and placebo arms.[25]

Two additional studies explored effect on survival and/or progression of administrations of other exogenous erythropoietin with higher hemoglobin targets.

In a randomized, placebo-controlled study using another Epoetin alfa product, conducted in 939 women with metastatic breast cancer, study drug dosing was titrated to attempt to maintain hemoglobin levels between 12 and 14 g/dL. At four months, death attributed to disease progression was higher (6% vs 3%) in women receiving Epoetin alfa. Overall mortality was significantly higher at 12 months in the

Epoetin alfa arm (See WARNINGS; Thrombotic Events and Increased Mortality).

In a randomized, placebo-controlled study using Epoetin beta, conducted in 351 patients with head and neck cancer, study drug was administered with the aim of achieving a hemoglobin level of 14 g/dL in women and 15 g/dL in men. Locoregional progression-free survival was significantly shorter (median PFS: 406 days Epoetin beta vs 745 days placebo, p = 0.04) in patients receiving Epoetin beta.[43]

There is insufficient information to establish whether use of Epoetin products, including PROCRIT®, have an adverse effect on time to tumor progression or progression-free survival.

These trials permitted or required dosing to achieve hemoglobin of greater than 12 g/dL. Until further information is available, the recommended target hemoglobin should not exceed 12 g/dL in men or women.

Surgery Patients

Thrombotic/Vascular Events

In perioperative clinical trials with orthopedic patients, the overall incidence of thrombotic/vascular events was similar in Epoetin alfa and placebo-treated patients who had a pre-treatment hemoglobin of > 10 g/dL to ≤ 13 g/dL. In patients with a hemoglobin of > 13 g/dL treated with 300 Units/kg of Epoetin alfa, the possibility that PROCRIT® treatment may be associated with an increased risk of postoperative thrombotic/vascular events cannot be excluded.[18-20,28]

In one study in which Epoetin alfa was administered in the perioperative period to patients undergoing coronary artery bypass graft surgery, there were 7 deaths in the group treated with Epoetin alfa (n = 126) and no deaths in the placebo-treated group (n = 56). Among the 7 deaths in the patients treated with Epoetin alfa, 4 were at the time of therapy (between study day 2 and 8). The 4 deaths at the time of therapy (3%) were associated with thrombotic/vascular events. A causative role of Epoetin alfa cannot be excluded (see WARNINGS).

Hypertension

Blood pressure may rise in the perioperative period in patients being treated with PROCRIT®. Therefore, blood pressure should be monitored carefully.

ADVERSE REACTIONS

Immunogenicity

As with all therapeutic proteins, there is the potential for immunogenicity. The observed incidence of antibody positivity in an assay may be influenced by several factors including assay methodology, sample handling, timing of sample collection, concomitant medications, and underlying disease. For these reasons, comparison of the incidence of antibodies to PROCRIT® with the incidence of antibodies to other products may be misleading.

A few cases of PRCA associated with antibodies with neutralizing activity have been reported in patients receiving PROCRIT® (see WARNINGS: PURE RED CELL APLASIA). These cases were observed in patients treated by either SC or IV routes of administration and occurred predominantly in CRF patients.

Chronic Renal Failure Patients

PROCRIT® is generally well-tolerated. The adverse events reported are frequent sequelae of CRF and are not necessarily attributable to PROCRIT® therapy. In double-blind, placebo-controlled studies involving over 300 patients with CRF, the events reported in greater than 5% of patients treated with PROCRIT® during the blinded phase were:

	Percent of Patients Reporting Event	
Event	Patients Treated With PROCRIT® (n = 200)	Placebo-treated Patients (n = 135)
Hypertension	24%	19%
Headache	16%	12%
Arthralgias	11%	6%
Nausea	11%	9%
Edema	9%	10%
Fatigue	9%	14%
Diarrhea	9%	6%
Vomiting	8%	5%
Chest Pain	7%	9%
Skin Reaction (Administration Site)	7%	12%
Asthenia	7%	12%
Dizziness	7%	13%
Clotted Access	7%	2%

Significant adverse events of concern in patients with CRF treated in double-blind, placebo-controlled trials occurred in the following percent of patients during the blinded phase of the studies:

Seizure	1.1%	1.1%
CVA/TIA	0.4%	0.6%
MI	0.4%	1.1%
Death	0	1.7%

In the US PROCRIT® studies in adult patients on dialysis (over 567 patients), the incidence (number of events per patient-year) of the most frequently reported adverse events were: hypertension (0.75), headache (0.40), tachycardia (0.31), nausea/vomiting (0.26), clotted vascular access (0.25), shortness of breath (0.14), hyperkalemia (0.11), and diarrhea (0.11). Other reported events occurred at a rate of less than 0.10 events per patient per year.

Events reported to have occurred within several hours of administration of PROCRIT® were rare, mild, and transient, and included injection site stinging in dialysis patients and flu-like symptoms such as arthralgias and myalgias.

In all studies analyzed to date, PROCRIT® administration was generally well-tolerated, irrespective of the route of administration.

Pediatric CRF Patients: In pediatric patients with CRF on dialysis, the pattern of most adverse events was similar to that found in adults. Additional adverse events reported during the double-blind phase in > 10% of pediatric patients in either treatment group were: abdominal pain, dialysis access complications including access infections and peritonitis in those receiving peritoneal dialysis, fever, upper respiratory infection, cough, pharyngitis, and constipation. The rates are similar between the treatment groups for each event.

Hypertension: Increases in blood pressure have been reported in clinical trials, often during the first 90 days of therapy. On occasion, hypertensive encephalopathy and seizures have been observed in patients with CRF treated with PROCRIT®. When data from all patients in the US phase 3 multicenter trial were analyzed, there was an apparent trend of more reports of hypertensive adverse events in patients on dialysis with a faster rate of rise of hematocrit (greater than 4 hematocrit points in any 2-week period). However, in a double-blind, placebo-controlled trial, hypertensive adverse events were not reported at an increased rate in the group treated with PROCRIT® (150 Units/kg TIW) relative to the placebo group.

Seizures: There have been 47 seizures in 1010 patients on dialysis treated with PROCRIT® in clinical trials, with an exposure of 986 patient-years for a rate of approximately 0.048 events per patient-year. However, there appeared to be a higher rate of seizures during the first 90 days of therapy (occurring in approximately 2.5% of patients) when compared to subsequent 90-day periods. The baseline incidence of seizures in the untreated dialysis population is difficult to determine; it appears to be in the range of 5% to 10% per patient-year.[39-41]

Thrombotic Events: In clinical trials where the maintenance hematocrit was 35 ± 3% on PROCRIT®, clotting of the vascular access (A-V shunt) has occurred at an annualized rate of about 0.25 events per patient-year, and other thrombotic events (eg, myocardial infarction, cerebral vascular accident, transient ischemic attack, and pulmonary embolism) occurred at a rate of 0.04 events per patient-year. In a separate study of 1111 untreated dialysis patients, clotting of the vascular access occurred at a rate of 0.50 events per patient-year. However, in CRF patients on hemodialysis who also had clinically evident ischemic heart disease or congestive heart failure, the risk of A-V shunt thrombosis was higher (39% vs 29%, $p < 0.001$), and myocardial infarctions, vascular ischemic events, and venous thrombosis were increased, in patients targeted to a hematocrit of 42 ± 3% compared to those maintained at 30 ± 3% (see WARNINGS).

In patients treated with commercial PROCRIT®, there have been rare reports of serious or unusual thrombo-embolic events including migratory thrombophlebitis, microvascular thrombosis, pulmonary embolus, and thrombosis of the retinal artery, and temporal and renal veins. A causal relationship has not been established.

Allergic Reactions: There have been no reports of serious allergic reactions or anaphylaxis associated with PROCRIT® administration during clinical trials. Skin rashes and urticaria have been observed rarely and when reported have generally been mild and transient in nature.

There have been rare reports of potentially serious allergic reactions including urticaria with associated respiratory symptoms or circumoral edema, or urticaria alone. Most reactions occurred in situations where a causal relationship could not be established. Symptoms recurred with rechallenge in a few instances, suggesting that allergic reactivity may occasionally be associated with PROCRIT® therapy. If an anaphylactoid reaction occurs, PROCRIT® should be immediately discontinued and appropriate therapy initiated.

Zidovudine-treated HIV-infected Patients

Adverse events reported in clinical trials with PROCRIT® in zidovudine-treated HIV-infected patients were consistent with the progression of HIV infection. In double-blind, placebo-controlled studies of 3 months duration involving approximately 300 zidovudine-treated HIV-infected patients, adverse events with an incidence of ≥ 10% in either patients treated with PROCRIT® or placebo-treated patients were:

	Percent of Patients Reporting Event	
Event	Patients Treated With PROCRIT® (n = 144)	Placebo-treated Patients (n = 153)
Pyrexia	38%	29%
Fatigue	25%	31%
Headache	19%	14%
Cough	18%	14%
Diarrhea	16%	18%
Rash	16%	8%
Congestion, Respiratory	15%	10%
Nausea	15%	12%
Shortness of Breath	14%	13%
Asthenia	11%	14%
Skin Reaction (Medication Site)	10%	7%
Dizziness	9%	10%

In the 297 patients studied, PROCRIT® was not associated with significant increases in opportunistic infections or mortality.[25] In 71 patients from this group treated with PROCRIT® at 150 Units/kg TIW, serum p24 antigen levels did not appear to increase.[27] Preliminary data showed no enhancement of HIV replication in infected cell lines *in vitro*.[25]

Peripheral white blood cell and platelet counts are unchanged following PROCRIT® therapy.

Allergic Reactions: Two zidovudine-treated HIV-infected patients had urticarial reactions within 48 hours of their first exposure to study medication. One patient was treated with PROCRIT® and one was treated with placebo (PROCRIT® vehicle alone). Both patients had positive immediate skin tests against their study medication with a negative saline control. The basis for this apparent pre-existing hypersensitivity to components of the PROCRIT® formulation is unknown, but may be related to HIV-induced immunosuppression or prior exposure to blood products.

Seizures: In double-blind and open-label trials of PROCRIT® in zidovudine-treated HIV-infected patients, 10 patients have experienced seizures.[25] In general, these seizures appear to be related to underlying pathology such as meningitis or cerebral neoplasms, not PROCRIT® therapy.

Cancer Patients on Chemotherapy

Adverse experiences reported in clinical trials with PROCRIT® administered TIW in cancer patients were consistent with the underlying disease state. In double-blind, placebo-controlled studies of up to 3 months duration involving 131 cancer patients, adverse events with an incidence > 10% in either patients treated with PROCRIT® or placebo-treated patients were as indicated below:

	Percent of Patients Reporting Event	
Event	Patients Treated With PROCRIT® (n = 63)	Placebo-treated Patients (n = 68)
Pyrexia	29%	19%
Diarrhea	21%*	7%
Nausea	17%*	32%
Vomiting	17%	15%
Edema	17%*	1%
Asthenia	13%	16%
Fatigue	13%	15%
Shortness of Breath	13%	9%
Paresthesia	11%	6%
Upper Respiratory Infection	11%	4%
Dizziness	5%	12%
Trunk Pain	3%*	16%

* Statistically significant

Although some statistically significant differences between patients being treated with PROCRIT® and placebo-treated patients were noted, the overall safety profile of PROCRIT® appeared to be consistent with the disease process of advanced cancer. During double-blind and subsequent open-label therapy in which patients (n = 72 for total exposure to PROCRIT®) were treated for up to 32 weeks with doses as high as 927 Units/kg, the adverse experience profile of PROCRIT® was consistent with the progression of advanced cancer.

Three hundred thirty-three (333) cancer patients enrolled in a placebo-controlled, double-blind trial utilizing Weekly dosing with PROCRIT® for up to 4 months were evaluable for adverse events. The incidence of adverse events was similar in both treatment and placebo arms.

Surgery Patients

Adverse events with an incidence of ≥ 10% are shown in the following table:

	Percent of Patients Reporting Event				
Event	Patients Treated With PROCRIT® 300 U/kg (n = 112)*	Patients Treated With PROCRIT® 100 U/kg (n = 101)*	Placebo-treated Patients (n = 103)*	Patients Treated With PROCRIT® 600 U/kg (n = 73)*	Patients Treated With PROCRIT® 300 U/kg (n = 72)*
Pyrexia	51%	50%	60%	47%	42%
Nausea	48%	43%	45%	45%	58%
Constipation	43%	42%	43%	51%	53%
Skin Reaction (Medication Site)	25%	19%	22%	26%	29%
Vomiting	22%	12%	14%	21%	29%
Skin Pain	18%	18%	17%	5%	4%
Pruritus	16%	16%	14%	14%	22%
Insomnia	13%	16%	13%	21%	18%
Headache	13%	11%	9%	10%	19%
Dizziness	12%	9%	12%	11%	21%
Urinary Tract Infection	12%	3%	11%	11%	8%
Hypertension	10%	11%	10%	5%	10%
Diarrhea	10%	7%	12%	10%	6%
Deep Venous Thrombosis	10%	3%	5%	0%	0%
Dyspepsia	9%	11%	6%	7%	8%
Anxiety	7%	2%	11%	11%	4%
Edema	6%	11%	8%	11%	7%

[a] Study including patients undergoing orthopedic surgery treated with PROCRIT® or placebo for 15 days

[b] Study including patients undergoing orthopedic surgery treated with PROCRIT® 600 Units/kg weekly x 4 or 300 Units/kg daily x 15

[c] Determined by clinical symptoms

Thrombotic/Vascular Events: In three double-blind, placebo-controlled orthopedic surgery studies, the rate of deep venous thrombosis (DVT) was similar among Epoetin alfa and placebo-treated patients in the recommended population of patients with a pretreatment hemoglobin of > 10 g/dL to ≤ 13 g/dL.[18,20,28] However, in 2 of 3 orthopedic surgery studies the overall rate (all pretreatment hemoglobin groups combined) of DVTs detected by postoperative ultrasonography and/or surveillance venography was higher in the group treated with Epoetin alfa than in the placebo-treated group (11% vs 6%). This finding was attributable to the difference in DVT rates observed in the subgroup of patients with pretreatment hemoglobin > 13 g/dL. However, the incidence of DVTs was within the range of that reported in the literature for orthopedic surgery patients.

In the orthopedic surgery study of patients with pretreatment hemoglobin of > 10 g/dL to ≤ 13 g/dL which compared two dosing regimens (600 Units/kg weekly x 4 and 300 Units/kg daily x 15), 4 subjects in the 600 Units/kg weekly PROCRIT® group (5%) and no subjects in the 300 Units/kg daily group had a thrombotic vascular event during the study period.[19]

In a study examining the use of Epoetin alfa in 182 patients scheduled for coronary artery bypass graft surgery, 23% of patients treated with Epoetin alfa and 29% treated with placebo experienced thrombotic/vascular events. There were 4 deaths among the Epoetin alfa-treated patients that were associated with a thrombotic/vascular event. A causative role of Epoetin alfa cannot be excluded (see WARNINGS).

OVERDOSAGE

The maximum amount of PROCRIT® that can be safely administered in single or multiple doses has not been determined. Doses of up to 1500 Units/kg TIW for 3 to 4 weeks have been administered to adults without any direct toxic effects of PROCRIT® itself.[6] Therapy with PROCRIT® can result in polycythemia if the hemoglobin is not carefully monitored and the dose appropriately adjusted. If the suggested target range is exceeded, PROCRIT® may be temporarily withheld until the hemoglobin returns to the suggested target range; PROCRIT® therapy may then be resumed using a lower dose (see DOSAGE AND ADMINISTRATION). If polycythemia is of concern, phlebotomy may be indicated to decrease the hemoglobin.

DOSAGE AND ADMINISTRATION

Chronic Renal Failure Patients

The recommended range for the starting dose of PROCRIT® is 50 to 100 Units/kg TIW for adult patients. The recommended starting dose for pediatric CRF patients on dialysis is 50 Units/kg TIW. The dose of PROCRIT® should be reduced as the hemoglobin approaches 12 g/dL or increases by more than 1 g/dL in any 2-week period. The dosage of PROCRIT® must be individualized to maintain the hemoglobin within the suggested target range. At the physician's discretion, the suggested target hemoglobin range may be expanded to achieve maximal patient benefit.

PROCRIT® may be given either as an IV or SC injection. In patients on hemodialysis, PROCRIT® usually has been administered as an IV bolus TIW. While the administration of PROCRIT® is independent of the dialysis procedure, PROCRIT® may be administered into the venous line at the end of the dialysis procedure to obviate the need for additional venous access. In adult patients with CRF not on dialysis, PROCRIT® may be given either as an IV or SC injection.

Patients who have been judged competent by their physicians to self-administer PROCRIT® without medical or other supervision may give themselves either an IV or SC injection. The table below provides general therapeutic guidelines for patients with CRF:

Starting Dose:	
Adults	50 to 100 Units/kg TIW; IV or SC
Pediatric Patients	50 Units/kg TIW; IV or SC
Reduce Dose When:	1. Hgb approaches 12 g/dL or, 2. Hgb increases >1 g/dL in any 2-week period
Increase Dose If:	Hgb does not increase by 2 g/dL after 8 weeks of therapy, and hgb is below suggested target range
Maintenance Dose:	Individually titrate
Suggested Target Hgb Range:	10 g/dL to 12 g/dL

During therapy, hematological parameters should be monitored regularly (see LABORATORY MONITORING).

Pretherapy Iron Evaluation: Prior to and during PROCRIT® therapy, the patient's iron stores, including transferrin saturation (serum iron divided by iron binding capacity) and serum ferritin, should be evaluated. Transferrin saturation should be at least 20%, and ferritin should be at least 100 ng/mL. Virtually all patients will eventually require supplemental iron to increase or maintain transferrin saturation to levels that will adequately support erythropoiesis stimulated by PROCRIT®.

Dose Adjustment: The dose should be adjusted for each patient to achieve and maintain a target hemoglobin not to exceed 12 g/dL.

Increases in dose should not be made more frequently than once a month. If the hemoglobin is increasing and approaching 12 g/dL, the dose should be reduced by approximately 25%. If the hemoglobin continues to increase, dose should be temporarily withheld until the hemoglobin begins to decrease, at which point therapy should be reinitiated at a dose approximately 25% below the previous dose. If the hemoglobin increases by more than 1 g/dL in a 2-week period, the dose should be decreased by approximately 25%.

If the increase in the hemoglobin is less than 1 g/dL over 4 weeks and iron stores are adequate (see PRECAUTIONS: LABORATORY MONITORING), the dose of PROCRIT® may be increased by approximately 25% of the previous dose. Further increases may be made at 4-week intervals until the specified hemoglobin is obtained.

Maintenance Dose: The maintenance dose must be individualized for each patient on dialysis. In the US phase 3 multicenter trial in patients on hemodialysis, the median maintenance dose was 75 Units/kg TIW, with a range from 12.5 to 525 Units/kg TIW. Almost 10% of the patients required a dose of 25 Units/kg, or less, and approximately

10% of the patients required more than 200 Units/kg TIW to maintain their hematocrit in the suggested target range. In pediatric hemodialysis and peritoneal dialysis patients, the median maintenance dose was 167 Units/kg/week (49 to 447 Units/kg per week) and 76 Units/kg per week (24 to 323 Units/kg/week) administered in divided doses (TIW or BIW), respectively to achieve the target range of 30% to 36%.

If the hemoglobin remains below, or falls below, the suggested target range, iron stores should be re-evaluated. If the transferrin saturation is less than 20%, supplemental iron should be administered. If the transferrin saturation is greater than 20%, the dose of PROCRIT® may be increased. Such dose increases should not be made more frequently than once a month, unless clinically indicated, as the response time of the hemoglobin to a dose increase can be 2 to 6 weeks. Hemoglobin should be measured twice weekly for 2 to 6 weeks following dose increases. In adult patients with CRF not on dialysis, the maintenance dose must also be individualized. PROCRIT® doses of 75 to 150 Units/kg/week have been shown to maintain hematocrits of 36% to 38% for up to 6 months.

Lack or Loss of Response: Over 95% of patients with CRF responded with clinically significant increases in hematocrit, and virtually all patients were transfusion-independent within approximately 2 months of initiation of PROCRIT® therapy.

If a patient fails to respond or maintain a response, other etiologies should be considered and evaluated as clinically indicated (see PRECAUTIONS: LACK OR LOSS OF RESPONSE).

Zidovudine-treated HIV-infected Patients

Prior to beginning PROCRIT®, it is recommended that the endogenous serum erythropoietin level be determined (prior to transfusion). Available evidence suggests that patients receiving zidovudine with endogenous serum erythropoietin levels > 500 mUnits/mL are unlikely to respond to therapy with PROCRIT®.

Starting Dose: For adult patients with serum erythropoietin levels ≤ 500 mUnits/mL who are receiving a dose of zidovudine ≤ 4200 mg/week, the recommended starting dose of PROCRIT® is 100 Units/kg as an IV or SC injection TIW for 8 weeks. For pediatric patients, see PRECAUTIONS: PEDIATRIC USE.

Increase Dose: During the dose adjustment phase of therapy, the hemoglobin should be monitored weekly. If the response is not satisfactory in terms of reducing transfusion requirements or increasing hemoglobin after 8 weeks of therapy, the dose of PROCRIT® can be increased by 50 to 100 Units/kg TIW. Response should be evaluated every 4 to 8 weeks thereafter and the dose adjusted accordingly by 50 to 100 Units/kg increments TIW. If patients have not responded satisfactorily to a PROCRIT® dose of 300 Units/kg TIW, it is unlikely that they will respond to higher doses of PROCRIT®.

Maintenance Dose: After attainment of the desired response (ie, reduced transfusion requirements or increased hemoglobin), the dose of PROCRIT® should be titrated to maintain the response based on factors such as variations in zidovudine dose and the presence of intercurrent infectious or inflammatory episodes. If the hemoglobin exceeds 13 g/dL,

the dose should be discontinued until the hemoglobin drops to 12 g/dL. The dose should be reduced by 25% when treatment is resumed and then titrated to maintain the desired hemoglobin.

Cancer Patients on Chemotherapy

Although no specific serum erythropoietin level can be stipulated above which patients would be unlikely to respond to PROCRIT® therapy, treatment of patients with grossly elevated serum erythropoietin levels (eg, > 200 mUnits/mL) is not recommended. The hemoglobin should be monitored on a weekly basis in patients receiving PROCRIT® therapy until hemoglobin becomes stable. The dose of PROCRIT® should be titrated to maintain the desired hemoglobin.

Two PROCRIT® dosing regimens may be used in adults; 150 Units/kg SC TIW or 40,000 Units SC Weekly. For pediatric patients, see PRECAUTIONS: PEDIATRIC USE.

TIW Dosing

Starting Dose:	
Adults	150 Units/kg SC TIW
Pediatric Patients	See PRECAUTIONS: Pediatric Use.
Reduce Dose by 25% when:	1. Hgb approaches 12 g/dL or, 2. Hgb increases > 1 g/dL in any 2-week period
Withhold Dose if:	Hgb exceeds 13 g/dL, until the hemoglobin falls to 12 g/dL, and restart dose at 25% below the previous dose
Increase Dose to 300 Units/kg TIW if:	response is not satisfactory [no reduction in transfusion requirements or rise in hemoglobin] after 8 weeks
Suggested Target Hgb Range:	10 g/dL to 12 g/dL

During therapy, hematological parameters should be monitored regularly (see PRECAUTIONS: Laboratory Monitoring).

Weekly Dosing

- The starting dose in adults is 40,000 Units SC Weekly. If after 4 weeks of therapy, the hemoglobin has not increased by ≥ 1 g/dL, in the absence of RBC transfusion, the PROCRIT® dose should be increased to 60,000 Units Weekly.
- If patients have not responded satisfactorily to a PROCRIT® dose of 60,000 Units Weekly after 4 weeks, it is unlikely that they will respond to higher doses of PROCRIT®.
- PROCRIT® should be withheld if the hemoglobin exceeds 13 g/dL and reinitiated with a 25% dose reduction when the hemoglobin is less than 12 g/dL.
- If PROCRIT® treatment produces a very rapid hemoglobin response (e.g., an increase of more than 1 g/dL in any 2-week period), the dose of PROCRIT® should be reduced by 25%.

Surgery Patients

Prior to initiating treatment with PROCRIT® a hemoglobin should be obtained to establish that it is > 10 to ≤ 13 g/dL.[18] The recommended dose of PROCRIT® is 300 Units/kg/day subcutaneously for 10 days before surgery, on the day of surgery, and for 4 days after surgery.

An alternate dose schedule is 600 Units/kg PROCRIT® subcutaneously in once weekly doses (21, 14, and 7 days before surgery) plus a fourth dose on the day of surgery.[19]

All patients should receive adequate iron supplementation. Iron supplementation should be initiated no later than the beginning of treatment with PROCRIT® and should continue throughout the course of therapy.

PREPARATION AND ADMINISTRATION OF PROCRIT®

1. Do not shake. It is not necessary to shake PROCRIT®. Prolonged vigorous shaking may denature any glycoprotein, rendering it biologically inactive.
2. Parenteral drug products should be inspected visually for particulate matter and discoloration prior to administration. Do not use any vials exhibiting particulate matter or discoloration.
3. Using aseptic techniques, attach a sterile needle to a sterile syringe. Remove the flip top from the vial containing PROCRIT®, and wipe the septum with a disinfectant. Insert the needle into the vial, and withdraw into the syringe an appropriate volume of solution.
4. **Single-dose:** 1 mL vial contains no preservative. Use one dose per vial; do not re-enter the vial. Discard unused portions.
   Multidose: 1 mL and 2 mL vials contain preservative. Store at 2° to 8°C after initial entry and between doses. Discard 21 days after initial entry.
5. Do not dilute or administer in conjunction with other drug solutions. However, at the time of SC administration, preservative-free PROCRIT® from single-use vials may be admixed in a syringe with bacteriostatic 0.9% sodium chloride injection, USP, with benzyl alcohol 0.9% (bacteriostatic saline) at a 1:1 ratio using aseptic technique. The benzyl alcohol in the bacteriostatic saline acts as a local anesthetic which may ameliorate SC injection site discomfort. Admixing is not necessary when using the multidose vials of PROCRIT® containing benzyl alcohol.

HOW SUPPLIED

PROCRIT®, containing Epoetin alfa, is available in vials containing color coded labels and caps.

1 mL **Single-Dose, Preservative-free** Solution

Each dosage form is supplied in the following packages:

Cartons containing six (6) **single-dose** vials:

2000 Units/mL (NDC 59676-302-01) (Purple)
3000 Units/mL (NDC 59676-303-01) (Magenta)
4000 Units/mL (NDC 59676-304-01) (Green)
10,000 Units/mL (NDC 59676-310-01) (Red)

Cartons containing four (4) **single-dose** vials:

40,000 Units/mL (NDC 59676-340-01) (Orange)

Trays containing twenty-five (25) **single-dose** vials:

2000 Units/mL (NDC 59676-302-02) (Purple)
3000 Units/mL (NDC 59676-303-02) (Magenta)
4000 Units/mL (NDC 59676-304-02) (Green)
10,000 Units/mL (NDC 59676-310-02) (Red)

2 mL **Multidose, Preserved** Solution

Cartons containing six (6) **multidose** vials:

10,000 Units/mL (NDC 59676-312-01) (Blue)

1 mL **Multidose, Preserved** Solution

Cartons containing six (6) **multidose** vials:

20,000 Units/mL (NDC 59676-320-01) (Lime)

STORAGE

Store at 2° to 8° C (36° to 46° F). Do not freeze or shake.

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Distributed by:
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ORTHO BIOTECH

Revised January 2005

638-10-979-4

Printed in U. S. A.

PROCRIT®
EPOETIN ALFA

INFORMATION FOR HOME DIALYSIS PATIENTS

What is PROCRIT® and how does it work?

PROCRIT® is a copy of human erythropoietin, a hormone produced primarily by healthy kidneys. PROCRIT® replaces the erythropoietin that the failed kidneys can no longer produce, and signals the bone marrow to make the oxygen-carrying red blood cells once again. PROCRIT® is produced in mammalian cells that have been genetically altered by the addition of a gene of the natural substance erythropoietin.

How should I take PROCRIT®?

In those situations where your doctor has determined that you, as a home dialysis patient, can self-administer PROCRIT®, you will receive instruction on how much PROCRIT® to use, how to inject it, how often you should inject it, and how you should dispose of the unused portions of each vial. You will be instructed to monitor your blood pressure carefully every day and to report any changes outside of the guidelines that your doctor has given you. When the number of red blood cells increases, your blood pressure can also increase, so your doctor may prescribe some new or additional blood pressure medication. Be sure to follow your doctor's orders. You may also be instructed to have certain laboratory tests, such as additional hematocrit or iron level measurements, done more frequently. You may be asked to report these tests to your doctor or dialysis center. Also, your doctor may prescribe additional iron for you to take. Be sure to comply with your doctor's orders. Continue to check your access, as your doctor or nurse has shown you, to make sure it is working. Be sure to let your health care professional know right away if there is a problem.

Allergy to PROCRIT®

Patients occasionally experience redness, swelling, or itching at the site of injection of PROCRIT®. This may indicate an allergy to the components of PROCRIT®, or it may indicate a local reaction. If you have a local reaction, consult your doctor. A potentially more serious reaction would be a generalized allergy to PROCRIT®, which could cause a rash over the whole body, shortness of breath, wheezing, reduction in blood pressure, fast pulse, or sweating. Severe cases of generalized allergy may be life-threatening. If you think you are having a generalized allergic reaction, stop taking PROCRIT® and notify a doctor or emergency medical personnel immediately.

How will I know if PROCRIT® is working?

The effectiveness of PROCRIT® is measured by the increase in hematocrit (the amount of red blood cells in the blood) that results from PROCRIT® therapy. The rise in hematocrit is not immediate. It usually takes about 2 to 6 weeks before the hematocrit starts to rise. The amount of time it takes, and the dose of PROCRIT® that is needed to make the hematocrit increase, varies from patient to patient.

What is the most important information I should know about PROCRIT® and CHRONIC RENAL FAILURE?

PROCRIT® has been prescribed for you by your doctor because you:

1. Have anemia due to your kidney disease.
2. Are able to dialyze at home.
3. Have been determined to be able to administer PROCRIT® without direct medical or other supervision.

A lack of energy or feeling of tiredness is the major symptom of anemia. Additional symptoms include shortness of breath, chest pain, and feeling cold all the time. The reason for these symptoms is that there is a lack of red blood cells. Red blood cells carry oxygen, which is important for all of the body's functions. When there are fewer red blood cells, the body does not get all the oxygen it needs.

Kidneys remove toxins from the blood; they also measure the amount of oxygen in the blood. If there is not enough oxygen, the kidneys will produce a hormone called erythropoietin. Erythropoietin is released into the bloodstream and travels to the bone marrow where red blood cells are made. Erythropoietin signals the bone marrow to make more oxygen-carrying red blood cells.

As the kidneys fail, they stop cleansing toxins from your blood. They also make less erythropoietin than they should. Therefore, the bone marrow does not receive a strong-enough signal to make the oxygen-carrying red blood cells. Fewer red blood cells are produced so the muscles, brain, and other parts of the body do not get the oxygen they need to function properly.

Most patients treated with PROCRIT® no longer need blood transfusions. However, certain medical conditions, or unexpected blood loss, may result in the need for a transfusion.

What do I need to know if I am giving myself PROCRIT® injections?

When you receive your PROCRIT® from the dialysis center, doctor's office or home dialysis supplier, always check to see that:

1. The name PROCRIT® appears on the carton and vial label.
2. You will be able to use PROCRIT® before the expiration date stamped on the package.

The PROCRIT® solution in the vial should always be clear and colorless. Do not use PROCRIT® if the contents of the vial appear discolored or cloudy, or if the vial appears to contain lumps, flakes, or particles. In addition, if the vial has been shaken vigorously, the solution may appear to be frothy and should not be used. Therefore, care should be taken not to shake the PROCRIT® vial vigorously before use. Unless you have been prescribed Multidose PROCRIT® (1 mL or 2 mL vials with a big "M" on the label, each containing a total of 20,000 Units of PROCRIT®), vials of PROCRIT® are for single use. Any unused portion of a vial should not be used. However, Multidose PROCRIT® may be used to inject

multiple doses as prescribed by your doctor, and may be stored in the refrigerator (but not the freezing compartment) between doses for up to 21 days, and can be used for multiple doses. Follow your doctor's or dialysis center's instructions on what to do with the used vials.

How should I store PROCRIT®?

PROCRIT® should be stored in the refrigerator, but not in the freezing compartment. Do not let the vial freeze and do not leave it in direct sunlight. Do not use a vial of PROCRIT® that has been frozen or after the expiration date that is stamped on the label. If you have any questions about the safety of a vial of PROCRIT® that has been subjected to temperature extremes, be sure to check with your dialysis unit staff.

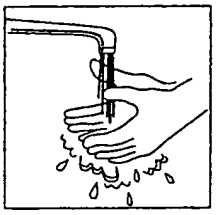
Always use the correct syringe.

Your doctor has instructed you on how to give yourself the correct dosage of PROCRIT®. This dosage will usually be measured in Units per milliliter or cc's. It is important to use a syringe that is marked in tenths of milliliters (for example, 0.2 mL or cc). Failure to use the proper syringe can lead to a mistake in dosage, and you may receive too much or too little PROCRIT®. Too little PROCRIT® may not be effective in increasing your hematocrit, and too much PROCRIT® may lead to a hematocrit that is too high. Only use disposable syringes and needles as they do not require sterilization; they should be used once and disposed of as instructed by your doctor.

IMPORTANT: TO HELP AVOID CONTAMINATION AND POSSIBLE INFECTION, FOLLOW THESE INSTRUCTIONS EXACTLY.

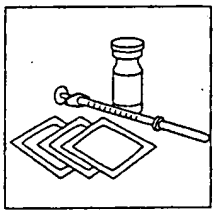
PREPARING THE DOSE

1. Wash your hands thoroughly with soap and water before preparing the medication.

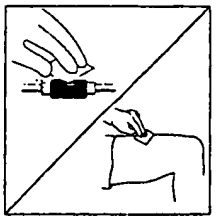


2. Check the date on the PROCRIT® vial to be sure that the drug has not expired.

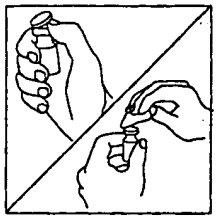
3. Remove the vial of PROCRIT® from the refrigerator and allow it to reach room temperature. Unless you are using a Multidose vial, each PROCRIT® vial is designed to be used only once. It is not necessary to shake PROCRIT®. Prolonged vigorous shaking may damage the product. Assemble the other supplies you will need for your injection.



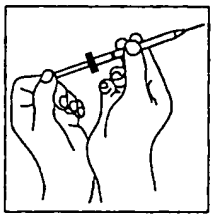
4. Hemodialysis patients should wipe off the venous port of the hemodialysis tubing with an antiseptic swab. Peritoneal dialysis patients should cleanse the skin with an antiseptic swab where the injection is to be made.



5. Flip off the protective cap but do not remove the gray rubber stopper. Wipe the top of the gray rubber stopper with an antiseptic swab.

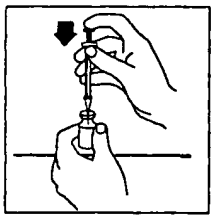


6. Using a syringe and needle designed for subcutaneous injection, draw air into the syringe by pulling back on the plunger. The amount of air should be equal to your PROCRIT® dose.



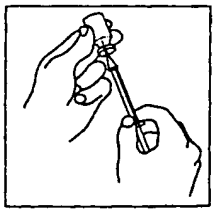
7. Carefully remove the needle cover. Put the needle through the gray rubber stopper of the PROCRIT® vial.

8. Push the plunger in to discharge air into the vial. The air injected into the vial will allow PROCRIT® to be easily withdrawn into the syringe.



9. Turn the vial and syringe upside down in one hand. Be sure the tip of the needle is in the PROCRIT® solution. Your other hand will be free to move the plunger. Draw back on the plunger slowly to draw the correct dose of PROCRIT® into the syringe.

10. Check for air bubbles. The air is harmless, but too large an air bubble will reduce the PROCRIT® dose. To remove air bubbles, gently tap the syringe to move the air bubbles to the top of the syringe, then use the plunger to push the solution and the air back into the vial. Then re-measure your correct dose of PROCRIT®.



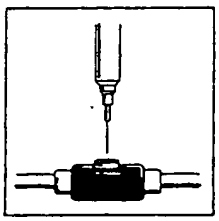
11. Double check your dose. Remove the needle from the vial. Do not lay the syringe down or allow the needle to touch anything.

INJECTING THE DOSE

Patients on home hemodialysis using the intravenous injection route:

1. Insert the needle of the syringe into the previously cleansed venous port and inject the PROCRIT®.

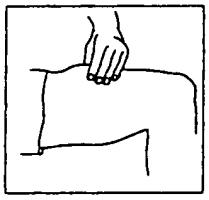
2. Remove the syringe and dispose of the whole unit. Use the disposable syringe only once. Dispose of syringes and needles as directed by your doctor, by following these simple steps:



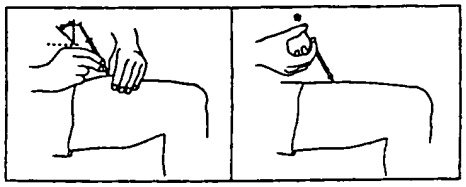
- Place all used needles and syringes in a hard plastic container with a screw-on-cap, or a metal container with a plastic lid, such as a coffee can properly labeled as to content. If a metal container is used, cut a small hole in the plastic lid and tape the lid to the metal container. If a hard-plastic container is used, always screw the cap on tightly after each use. When the container is full, tape around the cap or lid, and dispose of according to your doctor's instructions.
- Do not use glass or clear plastic containers, or any container that will be recycled or returned to a store.
- Always store the container out of the reach of children.
- Please check with your doctor, nurse, or pharmacist for other suggestions. There may be special state and local laws that they will discuss with you.

Patients on home peritoneal dialysis or home hemodialysis using the subcutaneous route:

1. With one hand, stabilize the previously cleansed skin by spreading it or by pinching up a large area with your free hand.



2. Hold the syringe with the other hand, as you would a pencil. Double check that the correct amount of PROCRIT® is in the syringe. Insert the needle straight into the skin (90 degree angle). Pull the plunger back slightly. If blood comes into the syringe, do not inject PROCRIT®, as the needle has entered a blood vessel; withdraw the syringe and inject at a different site. Inject the PROCRIT® by pushing the plunger all the way down.

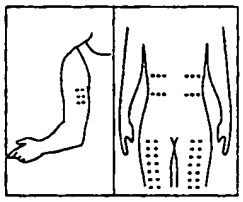


3. Hold an antiseptic swab near the needle and pull the needle straight out of the skin. Press the antiseptic swab over the injection site for several seconds.

4. Use the disposable syringe only once. Dispose of syringes and needles as directed by your doctor, by following these simple steps:

- Place all used needles and syringes in a hard plastic container with a screw-on-cap, or a metal container with a plastic lid, such as a coffee can properly labeled as to content. If a metal container is used, cut a small hole in the plastic lid and tape the lid to the metal container. If a hard-plastic container is used, always screw the cap on tightly after each use. When the container is full, tape around the cap or lid, and dispose of according to your doctor's instructions.
- Do not use glass or clear plastic containers, or any container that will be recycled or returned to a store.
- Always store the container out of the reach of children.
- Please check with your doctor, nurse, or pharmacist for other suggestions. There may be special state and local laws that they will discuss with you.

5. Always change the site for each injection as directed. Occasionally a problem may develop at the injection site. If you notice a lump, swelling, or bruising that doesn't go away, contact your doctor. You may wish to record the site just used so that you can keep track.



USAGE IN PREGNANCY

If you are pregnant or nursing a baby, consult your doctor before using PROCRIT®.

IMPORTANT NOTES

Since you are a home dialysis patient and your doctor allows you to self-administer PROCRIT®, please note the following:

1. Always follow the instructions of your doctor concerning the dosage and administration of PROCRIT®. Do not change the dose or instructions for administration of PROCRIT® without consulting your doctor.

2. Your doctor will tell you what to do if you miss a dose of PROCRIT®. Always keep a spare syringe and needle on hand.

3. Always consult your doctor if you notice anything unusual about your condition or your use of PROCRIT®.

Manufactured by:
Amgen Inc.
One Amgen Center Drive
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Printed in U.S.A.
Revised January 2005 638-10-979-4

 Printed in U.S.A. 2/05 08PCT1790R13

REPORTS

Recombinant Human Erythropoietin Therapy for Anemic Cancer Patients on Combination Chemotherapy

*Delvyn C. Case, Jr., Ronald M. Bukowski, Robert W. Carey, Ellioth H. Fishkin, David H. Henry, Robert J. Jacobson, Steven E. Jones, Allan M. Keller, John W. Kugler, Craig R. Nichols, Sydney E. Salmon, Richard T. Silver, Anna Maria Storniolo, Galen L. Wampler, Cathleen M. Dooley, Kay M. Larholt, Richard A. Nelson, Robert I. Abels**

Background: Patients with advanced cancer frequently experience clinically significant anemia, which is often exacerbated by myelosuppressive chemotherapy. Consistent with the anemia of chronic disease, studies have documented serum erythropoietin levels that are inappropriately low for the degree of anemia in cancer patients. Myelosuppressive chemotherapy impairs erythropoiesis, which may not fully recover between treatment cycles. Recombinant human erythropoietin (rHuEPO) has been used safely and effectively to treat anemia in AIDS patients receiving zidovudine (AZT) and in patients with chronic renal failure. *Purpose:* This study was designed to evaluate the clinical role of rHuEPO in reducing symptomatic anemia in patients with advanced cancer who were receiving myelosuppressive chemotherapy (excluding cisplatin). *Methods:* We studied 153 anemic cancer patients receiving cyclic combination chemotherapy in a prospective multicenter, double-blind, placebo-controlled trial. The patients were randomly assigned to receive either rHuEPO (150 U/kg) or placebo subcutaneously three times a week for a maximum of 12 weeks or until the hematocrit level increased to 38%-40%. If the hematocrit reached this target level before 12 weeks, the rHuEPO dose could be reduced to maintain the hematocrit at that level for the duration of the study. Response to rHuEPO therapy was assessed by measuring changes in hematocrit level, transfusion requirements, and quality of life. Quality-of-life assessment was based on patients' responses to questionnaires before and after the courses of therapy. *Results:* The increase in hematocrit in the rHuEPO-treated group compared with hematocrit in the placebo-treated group was statistically significant (P = .0001) as measured by percentage point of change from baseline to final evaluation, by an increase in hematocrit level of six percentage points or more unrelated to transfusion, and by a rise in hematocrit level to 38% or more unrelated to transfusion. There was a trend toward the reduction in mean units of blood transfused per patient during months 2 and 3 of therapy combined in rHuEPO-treated patients compared with placebo-treated patients (0.91 U versus 1.65 U; P = .056). In addition, rHuEPO-treated patients experienced a statistically significant improvement in energy level and ability to perform daily activities ($P \leq .05$). The two treatment groups showed no statistically significant differences in toxic effects except for increased incidence of diaphoresis ($P < .05$) and diarrhea (P = .05) in the rHuEPO-treated group. *Conclusions:* We conclude that rHuEPO is safe and effective for reversing anemia related to advanced cancer or to chemotherapy for cancer. [J Natl Cancer Inst 85:801–806, 1993]

Anemia is commonly observed in cancer patients and may be multifactorial in origin. One of the most common etiologies is the anemia of chronic disease, which is associated with neoplastic processes as well as chronic inflammation or infection. The anemia of chronic disease is characterized by erythroid hypoplasia of the bone marrow, a modest decrease in red blood cell survival, decreased bone marrow reutilization of iron, and inappropriately low erythropoietin (EPO) levels for the degree of anemia (*1*). Inappropriately low serum EPO levels for the degree of anemia in cancer patients have been documented (2) consistent with the model of anemia of chronic disease. Chemotherapy frequently exacerbates anemia in cancer patients. Periodic myelosuppressive chemotherapy impairs erythropoiesis, which may not fully recover by the time of the next cycle.

The symptomatology of anemia may contribute to the overall lack of well-being that patients with cancer may experience during their disease process. Such patients may require transfusions for palliation of the symptoms of anemia. Such transfusions carry clinically meaningful risks. It is estimated (*3,4*) that 20% of all blood transfusions will have some associated adverse effects (including fever, chills, rash, urticaria, and exposure to hepatitis). Because of the safety of recombinant human erythropoietin (rHuEPO) and its efficacy in the anemia of AIDS patients treated with zidovudine (AZT) (*5*) and in patients with chronic renal

*See "Notes" section following "References."

failure (6), we conducted three double-blind, placebo-controlled trials to evaluate the clinical utility of rHuEPO in anemic cancer patients receiving myelosuppressive chemotherapy (excluding cisplatin).

Patients and Methods

From October 1988 to June 1990, 157 patients were entered into three multicenter, double-blind, placebo-controlled clinical trials. The trials were designed to test whether the administration of rHuEPO to anemic cancer patients receiving cyclic combination chemotherapy would increase the hematocrit level, reduce the requirement for transfusion, and improve the quality of life.

The design of the three studies was essentially identical; an analysis of the demographics and other baseline data confirmed that the results could be pooled across protocols. Consequently, the results from the three studies have been pooled for this report.

Entry Criteria

For entry into the trial, anemia was defined as a hemoglobin concentration of 10.5 g/dL or less. All patients had a diagnosis of malignancy confirmed by biopsy specimen; primary myeloid malignancy and acute leukemia were excluded. All patients were receiving cyclic combination chemotherapy for a total of 5 days or less every 3-4 weeks. All patients had a performance status of 0, 1, 2, or 3 as defined by Miller et al. (7). Patients were required to be clinically stable for at least 1 month prior to study entry and to have an anticipated life expectancy of at least 3 months. All patients were more than 18 years old. Prior to study entry, we obtained a medical history, and a complete physical examination (including vital signs and weight) and a 12-lead electrocardiogram were performed for each patient. Laboratory values for eligibility were as follows: neutrophil count greater than 0.5×10^9 cells/L, platelet count greater than 75×10^9 cells/L, creatinine concentration less than 2.0 mg/dL, serum calcium level less than 12.0 mg/dL, serum folate, vitamin B12, serum iron, and total iron-binding capacity within normal limits or greater, reticulocyte count less than 3%, negative Coombs' test, and stool negative for occult blood. Pregnant women were not enrolled. Patients with known cerebral metastases, uncontrolled hypertension, acute illness within 7 days of study entry, experimental therapy within 30 days of study entry, or radiation or surgery within 30 days of study entry were also excluded.

Study Procedures

After the nature and potential consequences of therapy were explained, patients gave their informed consent to enter the study. The patients were randomly assigned to receive 150 U of rHuEPO per kilogram of body weight or placebo three times weekly by subcutaneous injection, with each dose separated from the next by at least 1 day. Baseline laboratory tests were conducted within 2 weeks prior to study entry. Baseline transfusion data were defined as the number of units of blood transfused per patient per month and as the proportion of patients who received transfusions during the 3-month baseline period prior to therapy. Dosing with the study medication was continued for 12 weeks or until the hematocrit normalized (reached the target range of 38%-40%). After the target hematocrit was attained, the rHuEPO dose could be reduced to maintain the hematocrit at 38%-40% for the duration of the study. We obtained complete blood counts weekly and blood chemistry panels monthly. Serum iron, total iron-binding capacity, ferritin, B12, and folate were measured prior to and at the end of the study. Levels of endogenous EPO were determined at the beginning of the study by radioimmunoassay (normal range, 4-26 mU/mL). Serum samples for the determination of antibodies against rHuEPO were obtained at entry and after completion of the study's 12-week double-blind phase or when a patient prematurely withdrew from the study. Decisions about red blood cell transfusion were at the discretion of the individual investigator. At the end of the double-blind portion of the trial, all patients (both those receiving placebo and those receiving rHuEPO) were allowed to enter an open-label phase, during which doses of rHuEPO as high as 300 U/kg, depending on the hematocrit response, were injected subcutaneously three times per week to achieve and maintain a hematocrit value between 38% and 40%. The results of the double-blind phase of this trial are reported here.

Quality-of-Life Determination and Physician Assessment of Efficacy

Performance status was measured before and after therapy following the criteria defined by Miller et al. (7). The quality-of-life assessment was based on patient response to a questionnaire (8), which asked each patient to evaluate his or her energy-level, ability to carry out daily activities, and overall quality of life during the past week by placing a vertical mark on a 100-mm line. The extremes of these lines represented the lowest and highest assessment of each category of well-being. The position of the mark on the bar represented, in millimeters, the patient's score for that particular assessment, with 0 being the worst and 100 being the best possible assessment for the previous week. This questionnaire was used before treatment began and at completion of therapy or early termination.

Drug Preparation and Supply

The rHuEPO and placebo were supplied by the Robert Wood Johnson Pharmaceutical Research Institute, Raritan, N.J. The rHuEPO (4000 U/mL and 10000 U/mL) was formulated as a sterile, buffered solution containing 2.5 mg/mL human serum albumin. The placebo contained an identical buffered vehicle containing 2.5 mg/mL of human serum albumin.

Statistical Procedures

Randomization was performed according to a computer-generated randomization code at the Robert Wood Johnson Pharmaceutical Research Institute.

Statistical inference for dichotomous variables formulated as 2×2 tables (e.g., sex by treatment group) was carried out using Fisher's Exact Test. The Extended-Mantel–Haenszel test with integer scores (9) was used for other types of discrete data such as Physician's Global Assessment. Two-sample t tests were used for between-group comparisons of means and paired t tests were used to test changes from baseline to final value. A linear model approach was used for inference on major efficacy variables such as change in hematocrit level, change in transfusion rates, and change in quality of life. The linear models were constructed with treatment group as the design factor and with various baseline measures, such as baseline hematocrit level, endogenous EPO level, bone marrow tumor involvement, and chemotherapy intensity (area under the curve for neutrophil count versus time), as covariants. All statistical tests of hypotheses were two sided and were carried out at the $\alpha = 0.05$ level.

Results

A total of 157 patients were enrolled in the three studies combined for this report (81 received rHuEPO and 76 received placebo). All patients were included in the safety analyses. Four patients (two receiving rHuEPO and two receiving placebo) were on therapy for less than 15 days and were excluded from efficacy analyses. The baseline characteristics, hematologic values, and transfusion history for all 157 patients are noted in Table 1. There were no statistically significant differences between treatment groups for these demographic characteristics or for baseline hematocrit levels, transfusion requirements, or endogenous EPO serum level. Most patients had low or appropriate serum EPO levels for the degree of anemia. Approximately 66% of the patients had endogenous EPO levels less than 150 mU/mL and fewer than 5% had values greater than 500 mU/mL. The distribution of cancer type in the rHuEPO and placebo treatment groups is presented in Table 2. There were no statistically significant between-group differences in the percentages of hematologic and non-hematologic tumors at baseline.

Table 1. Baseline hematologic values and transfusion history

Characteristic	rHuEPO	Placebo
No. of patients	81	76
Sex		
Male	33	29
Female	48	47
Median age, y (range)	64 (27-92)	64 (30-88)
Median neutrophil count, cells/μL	2925	3024
% patients transfused	54	51
Mean No. of red blood cell units transfused per patient over 3 months prior to study (range)	2.3 (0-25)	2.1 (0-16)
Mean hematocrit (%)	28.5 (18-40)	29.3 (23-40)
Mean endogenous EPO level, mU/mL (median)	143 (95.0)	151 (93.5)
Range	16-1262	16-1734

Table 2. Distribution of primary cancer type at baseline*

Type of malignancy	N (%)	N (%)
Hematologic	36 (46)	29 (39)
Breast	14 (18)	18 (24)
Gynecologic	9 (11)	8 (11)
Gastrointestinal	7 (9)	4 (5)
Lung (small cell and non-small cell)	6 (8)	9 (12)
Prostate	4 (5)	5 (7)
Head and neck	1 (1)	0 (0)
Other	1 (1)	0 (0)
Unknown primary	1 (1)	1 (1)

*Patients assessable for efficacy.

Hematocrit Response

Response to rHuEPO therapy related to changes in hematocrit was measured in three ways (Table 3): correction of anemia by a rise in hematocrit to 38% or more unrelated to transfusion; an increase of 6 percentage points or more in hematocrit unrelated to transfusion; and a change from baseline to final hematocrit level. As measured by these three criteria of response, patients receiving rHuEPO had a statistically significant improvement in the hematocrit level compared with the placebo group (P = .0001). Patients with hematologic malignancies or solid tumors responded equally well to rHuEPO treatment, and the presence of bone marrow metastases did not appear to reduce responsiveness to therapy.

Transfusion Requirements

Treatment with rHuEPO did not have a substantial effect on the transfusion rate when considering the full 12-week course of therapy. Further analysis showed no difference in transfusion requirements in the rHuEPO- and placebo-treated patients during the 1st month of therapy. However, there was a trend toward reduction in the percentage of rHuEPO-treated patients who received transfusions during months 2 and 3 of therapy and in the mean number of units of blood transfused during months 2 and 3 of therapy (Table 4). During months 2 and 3 combined, 28.6% of rHuEPO-treated patients received transfusions compared with 36.8% of placebo-treated patients; the mean number of units of blood transfused per patient during months 2 and 3 was 0.91 and 1.65, in the rHuEPO- and placebo-treated groups, respectively (P = .056).

Transfusion Practice and Intensity of Chemotherapy

It is unlikely that the results described above were related to differential transfusion practices, since the mean transfusion trigger (hematocrit level at which the patient was given a transfusion) in the two treatment groups was equivalent at about 24.5%. Since patients received a wide variety of different types of chemotherapy regimens, it appeared most appropriate to use surrogate markers for the intensity of chemotherapy. The surrogate markers chosen for the analysis of the intensity of chemotherapy (particularly for the intensity of chemotherapy-induced myelosuppression) were the effect on the absolute neutrophil count and the platelet count. Neutrophil analyses included comparison of the areas under the curve for neutrophil count versus time in the two treatment groups as well as comparison of the percentage of patients in each treatment group whose absolute neutrophil count fell below 1000 or 500 cells/μL. Platelet analyses included comparison in the change in platelet count from baseline to final determination in each treatment group, as well as comparison of the percentage of patients in each treatment

Table 3. Response of hematocrit to therapy

Response	rHuEPO (N = 79)	Placebo (N = 74)	P
Rise in hematocrit to ≥38% unrelated to transfusion	32 (40.5%)	3 (4.1%)	.0001
Six-percentage-point or more rise in hematocrit from baseline unrelated to transfusion	46 (58.2%)	10 (13.5%)	.0001
Mean percentage point change in hematocrit from prestudy to poststudy	6.9	1.0	.0001

Table 4. Mean number of transfused units per patient per month

	No. of patients	Patients transfused No.	Patients transfused %	Mean transfusion rate* (±SE)
rHuEPO				
Month 1	79	20	25.3	0.69 ± 0.16
Months 2 and 3	70	20	28.6	0.91 ± 0.27†
Placebo				
Month 1	74	20	27.0	0.71 ± 0.16
Months 2 and 3	68	25	36.8	1.65 ± 0.27†

*Least squares mean from linear analysis.
†Between-group difference P = .056.

group whose platelet counts fell below 50000 or 20000/μL (Table 5). These data suggest that the overall intensity of chemotherapy in the two treatment groups was equivalent and further suggest that the differences in hematocrit levels described above were not related to differential intensity of chemotherapy.

Effect of Baseline Serum EPO Level on Response to rHuEPO Therapy

A multivariate statistical analysis was used to assess the relationship between a patient's baseline endogenous EPO level and response to rHuEPO therapy. The results showed that baseline serum EPO levels were not associated with responsiveness to rHuEPO therapy in this patient population.

Quality-of-Life Assessment

Prestudy and poststudy quality-of-life assessments were available for 124 patients (rHuEPO = 63 and placebo = 61) (Fig. 1). The rHuEPO-treated population as a whole had a statistically significant ($P \leq .05$) increase in baseline-to-final evaluation for energy level and ability to perform daily activities, as well as a near statistically significant ($P = .086$) improvement for overall quality of life. No similar improvements in quality-of-life assessments were seen in placebo-treated patients. Moreover, the changes in quality-of-life scores were of somewhat greater magnitude in the rHuEPO-treated populations with an increase in hematocrit to 38% or more or an increase of six percentage points or more (both unrelated to transfusion) than in the rHuEPO-treated population as a whole (data not shown).

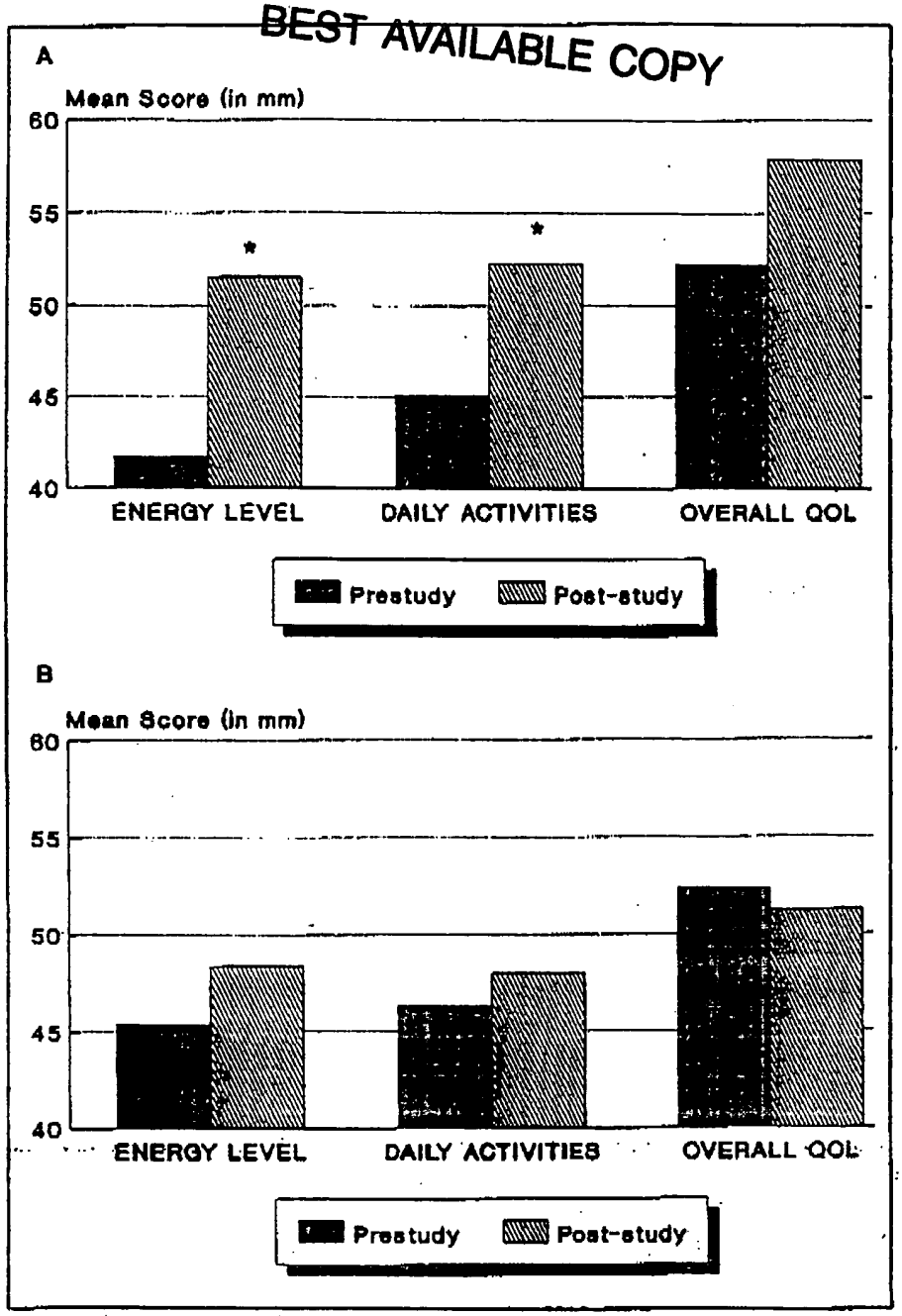

Fig. 1. Changes in quality-of-life measures for patients with prestudy and poststudy assessments. A) Patients who were treated with rHuEPO (N = 63). * = statistically significant improvement ($P \leq .05$) over prestudy value. B) Patients who received placebo (N = 61). QOL = quality of life.

Table 5. Changes in neutrophil or platelet counts during double-blinded therapy

Parameter	rHuEPO* (N = 79)	Placebo* (N = 74)
AUC† (cells × week/μL)	30203	34189
Neutrophils <1000/μL	51	48
Neutrophils <500/μL	32	23
Platelet counts/μL (% change from baseline to final value)	−39.0	−48.0
No. (%) of patients with platelet counts <50000/μL	18 (22.8)	17 (23.0)
No. (%) of patients with platelet counts <20000/μL	2 (2.5)	2 (2.7)

*No significant ($P > .005$) between-group differences.
†Area under the curve for neutrophil count versus time.

Safety

Overall, 78% of rHuEPO-treated patients completed the study versus 83% of the placebo-treated patients. Sixteen percent of the rHuEPO-treated patients discontinued study participation pre-

maturely because of an adverse experience, death, or disease progression versus 1.1% of the placebo-treated patients ($P>.05$).

There was no statistically significant difference in the incidence of any adverse experience in the rHuEPO-treated patients compared with placebo-treated patients except for diarrhea (22% on rHuEPO and 10% on placebo; $P = .05$) and diaphoresis (11% on rHuEPO and 1% on placebo; $P<.05$). Four patients treated with rHuEPO had hypertension reported as an adverse experience compared with two in the placebo group. Hypertension in the rHuEPO-treated patients tended to be more severe than in the placebo-treated patients, with the diastolic blood pressure in one of the rHuEPO-treated patients reaching 140 mm Hg. The hematocrit in this patient increased from 31% at baseline to 43% at the time the hypertension was reported (day 57). Four patients (two on rHuEPO and two on placebo) experienced seizures during therapy. The seizures in the rHuEPO-treated patients occurred in the context of a substantial increase in hematocrit and blood pressure. However, these patients also had structural abnormalities of the central nervous system (cerebral metastases and/or abnormal cells in the cerebrospinal fluid and increased cerebrospinal fluid protein) which may have contributed to their convulsive events. Thrombotic events (e.g., cerebrovascular accident) were noted in four patients treated with rHuEPO and four patients treated with placebo.

Discussion

The results of this double-blind, placebo-controlled study demonstrate that rHuEPO administered subcutaneously at a dose of 150 U/kg three times a week significantly ($P\leq.0001$) increased the hematocrit in anemic cancer patients who were treated with aggressive cyclic chemotherapy. The change in hematocrit from baseline to the end of the study was 5.9 percentage points greater in rHuEPO-treated patients than in placebo-treated patients. This stimulation of erythropoiesis in rHuEPO-treated patients was also reflected as an increase in the number of rHuEPO-treated patients whose anemias corrected when the patient achieved a hematocrit of 38% or more unrelated to transfusion or who responded to therapy with at least a six percentage point increase in hematocrit level unrelated to transfusion compared with placebo.

Data from other studies indicated that rHuEPO could effectively treat chemotherapy-induced anemia in patients with solid tumors (*10*) and hematologic malignancies with substantial bone marrow infiltration (*11*). Our data confirm that patients with hematologic or solid tumors responded equally well to rHuEPO treatment and that tumor infiltration of the bone marrow does not reduce responsiveness to therapy.

Overall, patients treated with rHuEPO therapy reported a statistically significant ($P\leq.05$) improvement in energy level and in the ability to perform daily activities and a trend toward improvement ($P = .083$) in the overall quality of life. This improvement is of potential significance, considering that these patients were not only in the late stages of cancer but were also enduring aggressive courses of chemotherapy. A subjective quality-of-life benefit associated with a response to rHuEPO therapy was also reported by Oster et al. (*11*) in patients with multiple myeloma.

Transfusion rates were not reduced during the 1st month of therapy in rHuEPO-treated patients compared with placebo-treated patients, but there was a clear trend toward reduced transfusion requirements in the rHuEPO-treated versus placebo-treated patients during months 2 and 3 of therapy. This lag in response was probably related to the time required for the stimulation of erythropoiesis to be reflected in decreased transfusion practice.

There was no statistically significant interaction between baseline endogenous EPO level and response to rHuEPO therapy observed in the patients studied. The absence of an interaction in our study is consistent with the findings of Miller et al. (2) for anemic cancer patients but differs from data from AZT-treated AIDS patients, where patients with lower serum EPO levels had a greater response to rHuEPO therapy than patients with higher baseline levels (*5*). The difference could be related to a temporary elevation in serum EPO levels in the cancer patients secondary to a recent course of intensive chemotherapy (*12-14*).

Therapy with rHuEPO was well tolerated in this patient population. While statistical comparison of the treatment groups indicated no increased cardiovascular risk to rHuEPO-treated patients, individual case histories suggest that meaningfully increasing hematocrit levels may occasionally be associated with an exacerbation of hypertension in this patient population. Based upon a comparable incidence of premature discontinuation due to adverse experiences, death, or disease progression and to a comparable treatment group distribution for prestudy and poststudy patient performance scores, there does not appear to be a deleterious effect of rHuEPO compared with placebo on the overall outcome in the treated population.

In conclusion, rHuEPO may be useful to correct the profound anemia that may be related to advanced cancer or to chemotherapy for cancer when other correctable causes of anemia (e.g., iron or folic acid deficiency) have been excluded.

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Notes

Affiliations of authors: D. C. Case., Jr., Maine Medical Center, Portland.

R. M. Bukowski, Cleveland Clinic/Cancer Center, Cleveland, Ohio.

R. W. Carey, Department of Medical Oncology, Massachusetts General Hospital, Boston.

E. H. Fishkin, Oncology Department, Elizabeth General Medical Center, Elizabeth, N.J.

D. H. Henry, Oncology/Hematology Department, Graduate Hospital, University of Pennsylvania, Philadelphia.

R. J. Jacobson, Division of Hematology, Georgetown University Hospital, Washington, D.C.

S. E. Jones, Department of Oncology, Sammons Cancer Center, Dallas, Tex.

A. M. Keller, Cancer Care Associates, Tulsa, Okla.

J. W. Kugler, Oncology/Hematology Associates of Central Illinois, Peoria.

C. R. Nichols, Section of Hematology, University Hospital, Indianapolis, Ind.

S. E. Salmon, Department of Oncology, The University of Arizona College of Medicine, Tucson.

R. T. Silver, Hematology/Oncology Department, New York Hospital, New York.

A. M. Stomiolo, Hematology/Oncology Department, University of California San Diego Medical Center, San Diego.

G. L. Wampler, Department of Hematology/Oncology, Medical College of Virginia, Richmond.

C. M. Dooley, K. M. Larholt, R. A. Nelson, R. I. Abels, The Robert Wood Johnson Pharmaceutical Research Institute, Raritan, N.J.

Correspondence to: Delvyn C. Case, Jr., M.D., Maine Medical Center, 22 Bramhall St., Portland, ME 04102.

Manuscript received August 10, 1992; revised January 27, 1993; accepted February 3, 1993.

Investigation of the Mechanism of Tamoxifen-Stimulated Breast Tumor Growth With Nonisomerizable Analogues of Tamoxifen and Metabolites

*Douglas M. Wolf, Susan M. Langan-Fahey, Christopher J. Parker, Raymond McCague, V. Craig Jordan**

Background: **The nonsteroidal antiestrogen tamoxifen (TAM) is the front-line endocrine treatment for breast cancer, but disease recurrence is common. Treatment failure may occur because tumors become insensitive to TAM. Alternatively, resistance may occur because tumors become stimulated rather than inhibited by TAM. TAM-stimulated growth of MCF-7 human breast tumors has been observed in athymic mice after prolonged treatment with TAM.** ***Purpose:*** **Our purpose was to examine the mechanism of treatment failure by determining whether TAM-stimulated tumors acquire the ability to excrete TAM and its antiestrogenic metabolites or to convert them to estrogenic compounds with weakened antiestrogenic activity.** ***Methods:*** **We used high-pressure liquid chromatography to quantitate TAM and its metabolites in serum and tumors from ovariectomized athymic mice and in MCF-7 cells grown in vitro. We treated tumor-bearing mice with subcutaneous sustained-release preparations of estradiol, TAM, or a nonisomerizable (fixed-ring) analogue and then assessed the activity of these compounds on TAM-inhibited parental MCF-7 tumors and on TAM-stimulated MCF-7 TAM tumors.** ***Results:*** **We found negligible differences in intratumoral TAM levels between TAM-inhibited parental MCF-7 tumors and TAM-stimulated MCF-7 TAM variants. We did not detect metabolite E (Met E), an estrogenic TAM metabolite, in serum or tumors. Using MCF-7 cells in vitro, we determined that the (Z) isomer of Met E, the form directly produced by TAM metabolism, must be present in the cell at a concentration of over 1000 ng/g to overcome growth inhibition by physiological levels of TAM and antiestrogenic metabolites, but the (E) isomer of Met E was effective at 10 ng/g. We reasoned that conversion of Met E from the (Z) (a weak estrogen) to (E) isomer (a potent estrogen) would be required if formation of Met E were responsible for TAM-stimulated growth. However, fixed-ring TAM, which can only form (Z) Met E, was shown to be as capable as TAM of initiating and maintaining antiestrogen-stimulated growth of MCF-7 tumors in athymic mice.** ***Conclusion:*** **Metabolism and isomerization of TAM to estrogenic compounds is not the mechanism of TAM-stimulated growth in our model.** ***Implication:*** **Other potential mechanisms for TAM-stimulated growth, such as estrogen receptor mutation, must be investigated so that effective strategies can be devised to control breast cancer once therapy fails. [J Natl Cancer Inst 85:806–812, 1993]**

Over the past two decades, the nonsteroidal antiestrogen tamoxifen (TAM) has become the standard endocrine treatment for all stages of breast cancer. It is used as a palliative therapy for women with advanced disease and as a postsurgical adjuvant treatment for women diagnosed with node-positive as well as node-negative disease. A recent overview analysis of clinical trials involving over 30000 women (*1*) showed that when used as an adjuvant, TAM is effective at prolonging both disease-free and overall survival. Unfortunately, the majority of node-positive patients (and a significant number of node-negative patients as well) will eventually experience disease

*See "Notes" section following "References."

Recombinant Human Erythropoietin Treatment in Cisplatin-Associated Anemia: A Randomized, Double-Blind Trial With Placebo

By Stefano Cascinu, Anna Fedeli, Elena Del Ferro, Stefano Luzi Fedeli, and Giuseppina Catalano

Purpose: **To evaluate the effect of exogenous recombinant human erythropoietin (rHuEPO) on the increase of hemoglobin levels and on the transfusion requirements in patients with cisplatin (CDDP)-induced anemia, we performed a double-blind randomized trial with placebo.**

Patients and Methods: **One hundred patients with CDDP-associated anemia (hemoglobin level < 90 g/L) were randomized to receive either placebo (saline solution) or rHuEPO (100 U/kg body weight subcutaneously) three times per week. The end points of this study were the increase in hemoglobin levels to greater than 100 g/L after 3, 6, and 9 weeks and the effect on transfusion requirements.**

Results: **Ninety-nine of 100 patients were assessable for response and toxicity. In the rHuEPO arm, mean hemoglobin levels were statistically significantly increased after the third, sixth, and ninth weeks of therapy (101.1 ± 9.0, 102.4 ± 6.6, and 105.1 ± 9.4 g/L, respectively) compared with the mean baseline value (86.3 ± 6.2 g/L). In the placebo arm, there were no increases in mean hemoglobin levels at the third, sixth, and ninth weeks (81.0 ± 5.2, 81.3 ± 9.2, and 81.2 ± 11 g/L, respectively) compared with the mean baseline value (87.3 ± 5.2 g/L). Furthermore only 20% of patients required blood transfusions in the rHuEPO arm versus 56% of patients in the placebo arm ($P = .01$), with a mean units of blood transfused per patient of 0.30 in the rHuEPO arm and 1.8 in the placebo arm ($P = .01$). Treatment was well tolerated, with no significant side effects.**

Conclusion: **CDDP-induced anemia is corrected by rHuEPO, which results in reduced blood transfusion requirements.**

J Clin Oncol 12:1058-1062. © 1994 by American Society of Clinical Oncology.

CISPLATIN (CDDP) treatment is one of the most common causes of chemotherapy-induced anemia. Approximately 40% of patients develop anemia during CDDP treatment and require packed RBC transfusions.[1,2] The anemia associated with CDDP therapy is a normochromic, normocytic, hypoproliferative anemia with a low reticulocyte count, similar to that seen in patients with chronic renal failure.[1,3] Although the etiology of this anemia is probably multifactorial, some studies have shown that, in this anemia, the inverse linear relationship between the concentrations of hemoglobin and of circulating erythropoietin that is observed in the other anemic states (iron deficiency, acute blood loss, and hemolysis) was not present, which is similar to the case of the anemia of chronic renal failure.[4-6]

In animal models of CDDP-associated anemia and in pilot studies of treatment with exogenous recombinant human erythropoietin (rHuEPO) has resulted in reversal of the anemia.[7-10] To evaluate the effect of erythropoietin on the increase of hemoglobin levels and on the transfusion requirements in patients with CDDP-induced anemia, we performed a double-blind placebo controlled trial.

From the Servizio di Oncologia, Ospedali Riuniti, Pesaro, Italy.
Submitted June 14, 1993; accepted January 3, 1994.
Address reprint requests to Stefano Cascinu, MD, Servizio di Oncologia, Ospedali Riuniti, P.le Cinelli 4, 61100 Pesaro, Italy.

0732-183X/94/1205-002753.00/0

PATIENTS AND METHODS

Patients

Cancer patients who were currently receiving a chemotherapy regimen containing CDDP were considered eligible for this study if they met the following criteria: hemoglobin levels greater than 110 g/L before chemotherapy; hemoglobin levels less than 90 g/L during treatment with CDDP; no severe symptoms or signs related to anemia that required blood transfusions; no previous chemotherapy; no previous radiation therapy to the pelvic, thoracic, or lumbar region; normochromic, normocytic anemia with a low reticulocyte count; absence of concomitant hemorrhage or hemolysis; no RBC transfusions in the 4 weeks before the current chemotherapy regimen; and adequate bone marrow, renal, hepatic, and cardiovascular functions before chemotherapy. Patients receiving androgen, antiandrogen or progestative therapy were excluded. CDDP chemotherapy was continued during the study. Informed consent was obtained from all study subjects and the study was approved by the ethical committee of our hospital.

Treatment Regimen

This study consisted of a two-arm, double-blind, placebo-controlled protocol that lasted 9 weeks. Subjects were randomly assigned to receive, by subcutaneous injection, either placebo (saline solution) or rHuEPO.

Randomization, using cards from a computer-generated list in sealed envelopes, was performed by a person not involved with the care or evaluation of the patients. The dose of rHuEPO was 100 U/kg body weight. If the hemoglobin level was greater than 120 g/L, rHuEPO was withheld until the hemoglobin level decreased to less than 100 g/L. Identical syringes with saline solution (placebo) or rHuEPO were prepared to maintain the double-blind design. Oral iron supplements were commenced if one of the following events occurred: (1) serum iron less than 50 μg/dL, (2) transferrin saturation less than 20%, or (3) serum ferritin less than 10 ng/mL.

Clinical and Laboratory Monitoring

Physical examinations were performed, vital signs recorded, and samples obtained for serum chemistry tests, serum ferritin, folate levels, hematology assessment, and urinalysis, at baseline and every week for the 9 weeks of the study. Chest x-rays and ECGs were obtained at baseline and at the end of the treatment.

Response Criteria

The end points of this study were an increase in hemoglobin concentration to greater than 100 g/L after 3, 6, and 9 weeks of therapy without transfusion, and the reduction of transfusion requirements. This level of hemoglobin (100 g/L) was chosen because patients with this degree of anemia generally have a good quality of life and do not need RBC transfusions. The trigger point for RBC transfusion in these patients was a hemoglobin level less than 80 g/L or the presence of symptoms such as dyspnea, tachycardia, or severe asthenia.

Evaluation of Adverse Effects

Evaluation of adverse effects, focused on hypertension or headache or other neurologic symptoms that have been linked to rHuEPO treatment in patients with end-stage renal disease or chronic renal failure, were noted at weekly physical examination for the duration of treatment. Any signs or symptoms of local irritation at the injection site, abnormal vital signs, or clinically significant abnormal laboratoristic findings were recorded for consideration as toxic effects or adverse reactions.

Statistical Analysis

Mean hemoglobin levels before and after treatment with rHuEPO or placebo were compared using a two-sided paired *t* test. Values are expressed as means ± SD. The association of pretreatment erythropoietin levels with response was determined using univariate logistic regression. A *P* value less than .05 was considered statistically significant.[11]

RESULTS

One hundred of 211 patients treated with a CDDP-containing regimen were enrolled onto this study. The remaining patients were considered not eligible because, although most of them were anemic, they did not present hemoglobin levels less than 90 g/L during chemotherapy. One patient in the placebo arm was not assessable for response because of rapid disease progression assessable after the second week of treatment.

Patient characteristics are listed in Table 1. Characteristics were balanced between the two arms. All patients had serum erythropoietin levels inappropriately low for the degree of anemia, with values ranging from 16 to 231 mU/L in the rHuEPO arm, and from 15 to 235 mU/L in the placebo arm. Only two patients for each arm presented an erythropoietin level greater than 200 mU/L. Furthermore, analysis of data showed that there was no correlation between baseline erythropoietin and hemoglobin levels in either the rHuEPO arm ($r = .25$, $P = .8$) or the placebo arm ($r = .2$, $P = .6$).

Table 1. Patient Characteristics on Day 1 of Study

Characteristic	rHuEPO	Placebo
Sex (no. of patients)		
Male	24	29
Female	26	21
Age, years		
Median	58	57
Range	44-72	45-68
Cancer		
Stomach	22	24
Ovary	12	10
Melanoma	2	2
Head and neck	7	8
Lung	4	4
Breast	3	2
Chemotherapeutic regimens		
CDDP 60 mg/m² every 2 weeks	3	2
CDDP (40 mg/m²) + 5FU (500 mg/m²) weekly	22	24
CDDP (60 mg/m²) + VP16 (100 mg/m²) every 3 weeks	6	6
CDDP (50 mg/m²) weekly	12	10
CDDP (100 mg/m²) + 5FU (1,000 mg/m²) every 3 weeks	7	8
Dose of CDDP (mg/m²)		
Median	180	200
Range	120-300	160-320
Hemoglobin level (g/L), mean ± SD	86.3 ± 6.2	87.3 ± 5.2
WBC count (× 10⁹/L), mean ± SD	8.0 ± 3.8	7.2 ± 4.0
Platelet count (× 10⁹/L), mean ± SD	276 ± 146	230 ± 113
Ferritin level (ng/mL), mean ± SD	680 ± 541	580 ± 459
Erythropoietin level (mU/L), mean ± SD	67.9 ± 66.6	49.3 ± 39.9

Abbreviations: 5FU, fluorouracil; VP16, etoposide.

Efficacy

Table 2 lists mean hemoglobin levels on day 1 and after 3, 6, and 9 weeks of therapy. In the rHuEPO arm, there was a statistically significant increase after 3, 6, and 9 weeks of therapy compared with baseline values, with all of the mean hemoglobin levels being greater than 100 g/L. In contrast, there were no increases in hemoglobin levels in the placebo arm. Furthermore, considering hemoglobin values in individual patients, we found that, in the rHuEPO arm, 29 of 50 patients (58%) at the third week, 37 of 50 (74%) at the sixth week, and 41 of 50 (82%) at the ninth week presented a hemoglobin level greater than 100 g/L, versus only one patient in the placebo arm, after the sixth week. None of the patients presented a hemoglobin level greater than 120 g/L during treatment with rHuEPO that required treatment discontinuation. The ability to respond to rHuEPO was independent

Table 2. Mean Hemoglobin Values on Day 1 and After 3, 6, and 9 Weeks of Therapy

Therapy	Baseline Hemoglobin Value	Hemoglobin Value After Weeks of Therapy		
		3	6	9
rHuEPO	86.3 ± 6.2	101.1 ± 9.0*	102.4 ± 6.6*	105.1 ± 9.4*
Placebo	87.3 ± 5.2	81.0 ± 5.2	81.3 ± 9.2	81.2 ± 11

*$P \leq .01$ v baseline value.

of pretreatment erythropoietin levels ($P = .27$, univariate logistic regression model).

Regarding transfusion requirements, an advantage for rHuEPO treatment was observed. In fact, in the rHuEPO arm, only 20% of patients required blood transfusions versus 56% of patients in the placebo arm ($P = .01$), with a mean units of blood transfused per patient of 0.30 in the rHuEPO arm and 1.8 in the placebo arm ($P = .01$) (Table 3).

Five patients (10%) in the rHuEPO arm developed iron deficiency (serum iron < 50 μg/dL and transferrin saturation < 20%) and required iron supplementation after the third week of treatment.

Adverse Effects

Treatment was well tolerated. No patient was removed from the study because of rHuEPO-related toxicity. None of the patients developed hypertension, seizures, or thrombohemorrhagic complications.

DISCUSSION

Anemia is a common adverse effect following CDDP chemotherapy. It can adversely affect the patients' quality of life, and RBC transfusions are often required.[1,2] Although risks from blood transfusion are rapidly decreasing, 20% of all blood transfusions are still associated with at least some adverse reaction.[12,13] A further problem may be represented by religious beliefs that do not allow blood transfusions. Considering all of these data, an effective treatment for CDDP-induced anemia could improve the quality of life for these patients, and reduce the related anemia symptoms, the risks associated with blood transfusions, and even the ethical or psychologic impact.

Although the mechanism of CDDP-induced anemia is not well known, it appears that inadequate erythropoietin response, shown by the absence of the linear relationship between the concentration of hemoglobin and of circulating erythropoietin, is important in the development of this anemia.[3-6,14] This inadequate response was thought to be due to CDDP-associated nephrotoxicity.[6] However, in previous studies, renal function was not impaired by CDDP treatment, although subclinical nephrotoxicity could not be excluded.[8,10] Moreover, the erythropoietin response to anemia was similar in patients who received chemotherapy whether or not the treatment included CDDP. This suggests that chemotherapy may have an effect on the erythropoietin response to anemia that is independent of therapy-induced nephrotoxicity.

Also in our study, we found erythropoietin levels that were inappropriately low for the degree of anemia, as well as the lack of a correlation between erythropoietin and hemoglobin levels without signs of clinical nephrotoxicity.

Recently, on the basis of results obtained in animal models, some pilot studies were performed and obtained interesting results on rHuEPO treatment of CDDP-induced anemia.[8-10] To confirm these preliminary results, we performed the present randomized, double-blind, placebo-controlled trial.

In this study, the rHuEPO route of administration was chosen on the basis of data available in the literature and of our previous experience. rHuEPO subcutaneous injections allow a slow release from subcutaneous depots, providing lower but more sustained plasma levels than intravenous injections. In fact, the pharmacokinetics of intravenously administered rHuEPO are characterized by brief peaks in plasma levels due to the relatively small distribution volume, about the same as the plasma volume, and the short half-life of approximately 6 to 8 hours.[15,16] For these reasons, subcutaneous administration can be advantageous, because even lower doses may be sufficient for a certain erythropoietic effect. Furthermore, a subcutaneous route of rHuEPO administration was shown to be effective and safe in the treatment of anemia associated with chronic renal failure, myeloma, and other hematologic diseases, and it can be administered on an outpatient basis.[17-19]

A lower dose than that usually administered was chosen on the basis of the data reported here, of preclinical findings, and of our previous pilot study. Low doses of rHuEPO were shown to be effective in reversing the CDDP-induced anemia, whereas higher doses were required for the treatment of anemia induced by other cytotoxic drugs.[7] Finally, we demonstrated that rHuEPO at the dose of 100 U/kg body weight was able to maintain

Table 3. Hemotransfusion Requirements

Parameter	Treatment Group		P
	rHuEPO (N = 50)	Placebo (N = 49)	
Patients transfused (%)	20	58	.01
Mean units of blood transfused per patient	0.30	1.8	.01

hemoglobin levels greater than 100 g/L in 17 of 20 patients with CDDP-associated anemia.[10]

In the present randomized, double-blind trial, we confirmed the efficacy and safety of subcutaneous rHuEPO in the treatment of CDDP-induced anemia, considering either the increase in hemoglobin levels or the transfusion requirements. As reported in other clinical trials, we did not find a significant relationship between the response to rHuEPO and baseline endogenous erythropoietin levels.[8-10] However, similarly to previous studies, in our patients, pretreatment serum erythropoietin levels were generally low, with only two patients showing baseline erythropoietin levels greater than 200 mU/L. It is well known that, within the range of values observed in CDDP-associated anemia, pretreatment serum erythropoietin level does not seem to represent a factor related to responsiveness to exogenous rHuEPO.[8-10]

An interesting finding arising from our study is that in the rHuEPO arm, despite the similar mean hemoglobin values at the third, sixth, and ninth weeks of therapy (101.1 ± 9, 102.4 ± 6.6, and 105.1 ± 9.4 g/L, respectively), a greater number of patients reached the target hemoglobin level (100 g/L) with the continuation of treatment. Fifty-eight percent of patients achieved the target hemoglobin level at the third week, 74% at the sixth week, and 82% at the ninth week. These data seem to suggest that the continuation of rHuEPO treatment, at least using this dose and route of administration, can determine the achievement of the target hemoglobin level, even in initially nonresponding patients, without, at the same time, increasing hemoglobin levels too much in early-responding patients. Indeed, the presence of late responses to rHuEPO has been observed previously in anemic patients with end-stage renal disease and in the treatment of anemia associated with multiple myeloma.[17,18]

Furthermore, Abels et al[20] reported analog findings in their study on the treatment of CDDP-associated anemia. They found that the administration of rHuEPO 150 U/kg three times per week resulted in a progressive increase in the number of patients presenting with an increasing hematocrit value after 4 to 5 weeks of therapy. Considering its probable clinical importance, the possibility of obtaining an amelioration of the anemic state with the continuation of rHuEPO therapy in patients who have not responded in the first weeks of treatment should be carefully evaluated in further studies.

Despite the obvious benefits of rHuEPO therapy, a problem could be presented by the cost of treatment. Taking into account that rHuEPO is a relatively expensive molecule, a formal cost-benefit analysis should be performed in a future controlled trial on the treatment of CDDP-associated anemia to evaluate if this treatment is cost-effective, as found in the treatment of patients with chronic renal failure or hematologic disorders.[17,19]

In conclusion, the results of this double-blind randomized study, which confirm the preliminary data obtained in phase I/II clinical trials, show that rHuEPO may be a useful therapy to palliate the significant anemia often associated with CDDP chemotherapy, because it can increase hemoglobin values and reduce the need for hemotransfusions.

ACKNOWLEDGMENT

We thank Professor Clara Viola of the Department of Mathematics, Ancona University (Ancona, Italy) for statistical analyses.

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